Application Note

Environmental
Water Testing

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Haloacetic Acid Analysis Using Ion Chromatography (IC) Followed by Triple Quadrupole Mass Spectrometry (MS) Detection

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Abstract

This application note presents a direct injection method for the analysis of nine haloacetic acids (HAAs) using the Metrohm ion chromatograph (IC) and the Agilent 6470 triple quadrupole (TQ) LC/MS. United Sates (US) Environmental Protection Agency (EPA) Method 557 performance criteria were demonstrated, and low reporting levels were achieved. Spike recoveries in synthetic matrix were 86 to 104% and ranged from 72 to 121% in finished drinking water. All calibration curves were linear (R^2 >0.99) and showed excellent accuracy.

Introduction

HAAs are disinfection by-products generated when natural water containing dissolved organic matter is chlorinated. Chlorination is a common treatment process that kills bacteria, viruses, and other microbes in water, making it safe for potable use. The US EPA has determined that long-term exposure to five HAAs (HAA5, bold compounds in Table 1) can increase health risks.[1] A maximum contaminant level (MCL) of 60 µg/L total has been set as the National Primary Drinking Water Regulation (NPDWR) in the US.[1]

The most common method for analysis in the US is EPA Method 552.2, which involves extracting the HAAs from water and derivatizing to the methyl ester followed by gas chromatography (GC)-electron capture detection (ECD) analysis.[2] While low detection levels can be achieved using GC-ECD, the method is labor-intensive and can take several days to complete. The derivatization steps required by EPA Method 552.2 can add uncertainty to surrogate recoveries. Alternatively, EPA Method 557 is a direct injection method[3] that greatly reduces sample preparation time and adds sensitivity due to its improved accuracy.

EPA Method 557 uses ion chromatography (IC) separation, followed by triple quadrupole (TQ) mass spectrometry (MS) detection. The technique is more selective than GC-ECD, because specific mass transitions are monitored for each compound in a process called multiple reaction monitoring (MRM). MRM selectively allows the use of mass labeled internal standards (Table 1) to monitor and correct for variations in method performance. Another consideration inherent to IC anionic separations is the presence of anions (i.e., Cl^-, NO_3^-, etc.) commonly found in water, which are monitored using conductivity detection before injection onto the MS.

Table 1. HAA9 and internal standards

HAA	CAS No.	Internal Standard
Bromochloroacetic Acid (BCAA)	5589-96-3	
Bromodichloroacetic Acid (BDCAA)	7113-314-7	
Chlorodibromoacetic Acid (CDBAA)	5278-95-5	
Dibromoacetic Acid (DBAA)	631-64-1	
Dichloroacetic Acid (DCAA)	79-43-6	Dichloroacetic acid-2-^{13}C
Monobromoacetic Acid (MBAA)	79-08-3	Monobromoacetic acid-1-^{13}C
Monochloroacetic Acid (MCAA)	79-11-8	Monochloroacetic acid-2-^{13}C
Tribromoacetic Acid (TBAA)	75-96-7	
Trichloroacetic Acid (TCAA)	76-03-9	Trichloroacetic acid-2-^{13}C

The 6470B TQ LC/MS was selected for EPA Method 557 verification because of its sensitive and robust performance. This application note demonstrates the method requirements for all nine HAA compounds (Table 1). Low detection levels were achieved in a 40-minute run. Analysis of HAAs using the Agilent 6490 TQ LC/MS[4] and the 6470 TQ LC/MS[5] have been described previously.

Experimental

Sample preparation

Water samples were collected, preserved with 100 mg/L ammonium chloride, and stored at 4 °C until analysis. Before injection, 10 mL of sample was aliquoted into polypropylene tubes (Metrohm part number 6.2743.057) and 50 µL of labeled internal standard (IS) mix was added for a final concentration of 10 ng/mL. The tubes were capped and vortexed vigorously. Calibration standards and required QC matrix samples were prepared in the same way.

IC/TQ analysis and instrumentation

IC/TQ analysis was carried out with a Metrohm 940 Professional IC Vario TWO/SeS/PP coupled to a 6470B TQ LC/MS. The IC was equipped with a Metrosep A SUPP 7 250 mm by 4 mm column. The IC conditions and gradient are provided in Table 2 and Figure 1, respectively. The chemical suppressor is automatically regenerated with 1 M/5% methanol nitric acid solution delivered by the 800 Dosino (Metrohm part number 6.5330.1900) and then rinsed with reagent water delivered by the peristaltic pump on the 940 IC.

Table 2. IC parameters.

Parameter	Value
Injection Loop	100 µL
Column	Metrosep A supp 7 250/4.0
Mobile Phase A	75 mMol KOH + 5% methanol
Mobile Phase B	Ultrapure water
Suppressor Regenerant	HNO_3 = 1 mol/L + 5% methanol
Suppressor Rinse	Ultrapure water
Run Time	40 minutes

The 6470B TQ LC/MS was operated in the dynamic MRM mode. Two transitions per compound were monitored when available. Flow was diverted away from the MS at the beginning and end of the analysis. The MS ion source parameters and compound-specific MRM parameters are provided in Tables 3 and 4, respectively.

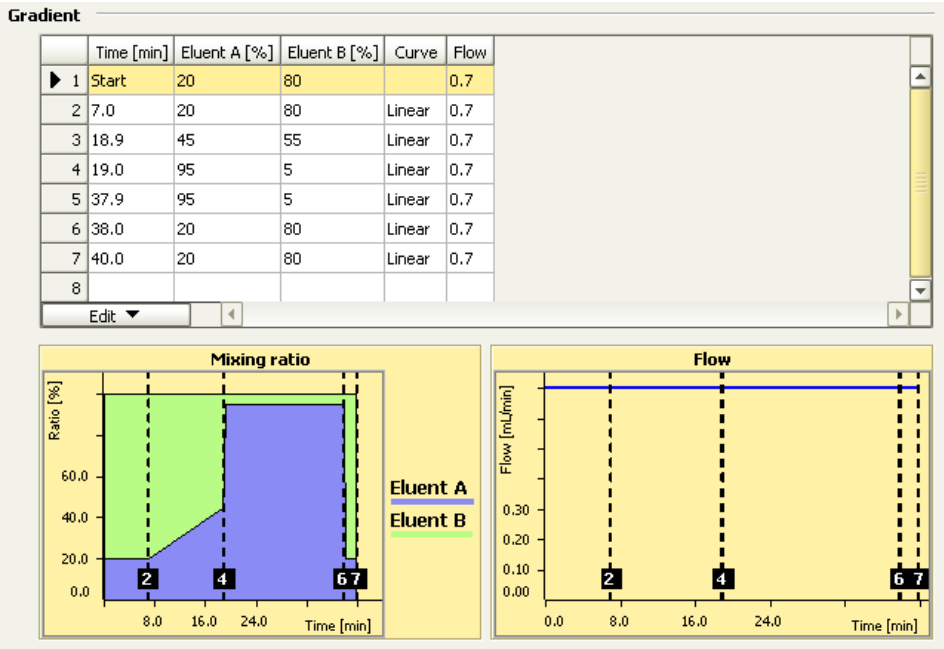

Figure 1. IC gradient.

Table 3. Ion source parameters.

Parameter	Value
Drying Gas Temperature	150 °C
Drying Gas Flow	12 L/min
Nebulizer Pressure	55 psi
Sheath Gas Heater Temperature	245 °C
Sheath Gas Flow	12 L/min
Capillary Voltage	3,500 V
Nozzle	2,000 V

The IC and 6470B TQ LC/MS were connected through a remote box. The MagicNet (version 3.3) determination series and the Agilent MassHunter software worklist were operated in parallel with MagicNet initiating a remote start 2.5 minutes into the IC separation. Conductivity chromatograms were available through MagicNet; MassHunter software (version 10.1) was used for quantification.

As noted in EPA Method 557, precursor ions can differ based on the electrospray ionization (ESI) interface design.[3] In this case, the highest abundance precursors of BDCAA, CDBAA, TCAA, and TBAA corresponded to a loss of a carboxyl group and were selected for quantitation.

Method evaluation samples

Reagent water samples containing the preservative and internal standards were used to monitor system background and carry over. Each sample set started and ended with a blank after the highest calibration point.

Table 4. HAA-specific 6470B TQ LC/MS MRM parameters.

Cpd Name	Precursor Ion	Product Ion	Frag (V)	CE (V)	Cell Acc (V)	Polarity
BCAA	173	129	60	10	2	Negative
BCAA	173	80.9	60	25	2	Negative
BDCAA	163	81	100	10	2	Negative
BDCAA	163	78.9	100	10	2	Negative
CDBAA	206.9	81	100	15	2	Negative
CDBAA	206.7	79.1	100	15	2	Negative
DBAA	216.8	173	60	10	2	Negative
DBAA	216.8	78.9	60	35	2	Negative
DCAA	127	83	80	10	2	Negative
DCAA	127	34.8	80	25	2	Negative
DCAA-2-^{13}C	128	84	80	10	2	Negative
MBAA	137	79	60	10	2	Negative
MBAA-1-^{13}C	138	79.1	60	10	2	Negative
MCAA	93	35	80	10	2	Negative
MCAA-2-^{13}C	94	35	80	10	2	Negative
TBAA	250.9	78.9	50	25	7	Negative
TCAA	161	117	40	5	2	Negative
TCAA	117	34.9	100	15	2	Negative
TCAA-2-^{13}C	118	34.9	80	15	2	Negative

Calibration was performed with a seven-point curve covering 0.5 to 40 ng/mL using the required internal standards (IS). IS areas of samples, quality control (QC) samples, and continuing calibration (CC) samples were monitored and required to be within ±50% of the initial calibration.

The laboratory fortified synthetic sample matrix (LFSSM) was prepared as described in EPA Method 557. In short, it contained 100 mg/L of ammonium chloride, 20 mg/L of nitrate, 150 mg/L of bicarbonate, 250 mg/L of chloride, and 250 mg/L of sulfate. This synthetic matrix was fortified with 10 µg/L of HAAs and injected at the beginning of every sample batch. Both HAA recovery (±30%) and IS areas (±50%) were monitored to ensure that the anions did not cause matrix suppression. The LFSSM was also used to determine method precision and accuracy. Seven replicate injections fortified at 10 µg/L were analyzed in conjunction with seven injections of the laboratory fortified blanks.

Method reporting level confirmation was accomplished by fortifying seven replicate reagent water samples containing the preservative and IS. Most HAAs were tested for prediction interval of results (PIR) at 0.5 ng/mL. TBAA was spiked higher (2 ng/mL) for the MRL calculation.

Samples analyzed

Tap water from Pennsylvania and Delaware was collected and divided into two aliquots. One served as the laboratory fortified sample matrix (LFSM) after being spiked with 10 ng/L HAA. Spike recoveries (±30%) were evaluated after correction for the native levels of HAAs. The Pennsylvania water was collected before and after an in-home filtration system.

Results and discussion

Demonstration of capability

The IC method provided good separation of all the HAAs. A chromatogram at the minimum report level (0.5 ng/mL, except TBAA, which is 2 ng/mL) is shown in Figure 2.

Reagent blanks containing the preservative and ISs were consistently clean at the required <1/3 MRL. Only MCAA and DCAA showed minimal peaks, likely from the labeled IS. In all cases, the levels were well below the required level.

The 6470 TQ LC/MS showed excellent linearity across the tested range. All fits were linear and weighting (1/x) was used for MCAA only. R^2 values were >0.99 and accuracy for all points on the curve was within ±20%.

Replicate analysis of the LFSSM showed good precision and accuracy. The relative standard deviation was highest for TBAA (20%). The relative standard deviations of all other compounds were 10 to 12%. Accuracy values were 86 to 104%.

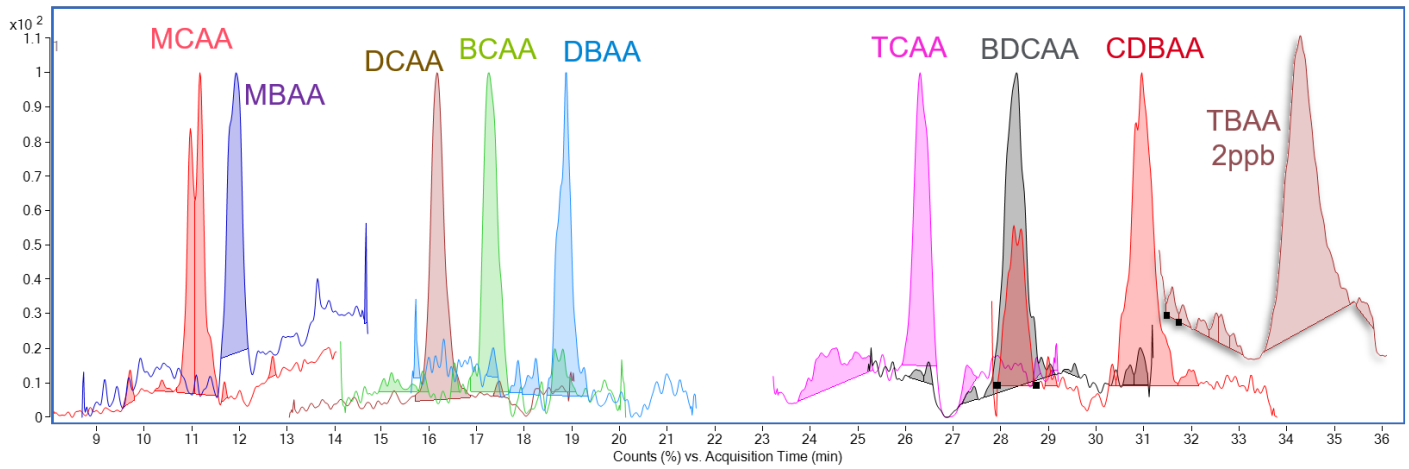

Figure 2. IC/MS chromatogram showing good separation of the HAAs at 0.5 µg/L (except for TBAA, which was at 2 µg/L).

Sample results

Figure 3 shows the conductivity chromatogram for the Pennsylvania tap water samples. The anions Cl^-, NO_3^-, and SO_4^- were present. The carbonate was removed by the Metrohm carbonate suppressor.

Spike recovery was within method limits and ranged from 72 to 121% across the water types. The native samples contained HAAs, but they were all below the MCL of 60 µg/L, with the highest measured at 11 µg/L total. If all HAA9 were included, the maximum level observed was 19 µg/L. The tap water tested before and after the in-home filtration system showed a reduction in total HAA9 from 19 µg/L prefiltration to 2.6 µg/L post filtration.

Conclusion

The IC/TQ method demonstrated good measurement performance for all HAA9 in both synthetic and tap water. The Metrohm IC provided excellent separation of the native HAAs and the 6470 TQ LC/MS provided selective, sensitive, and robust quantification of HAAs in water containing parts per million (ppm) levels of anions. Total run time was reduced to 40 minutes from 55 minutes as described in EPA Method 557, while maintaining all the performance-based criteria for spike recoveries and IS area deviations. Minimum reporting levels were 0.5 µg/L for all compounds, except for TBAA, which was 2 µg/L. When applied to real water samples, several HAAs were detected at low levels, demonstrating the performance of the method.

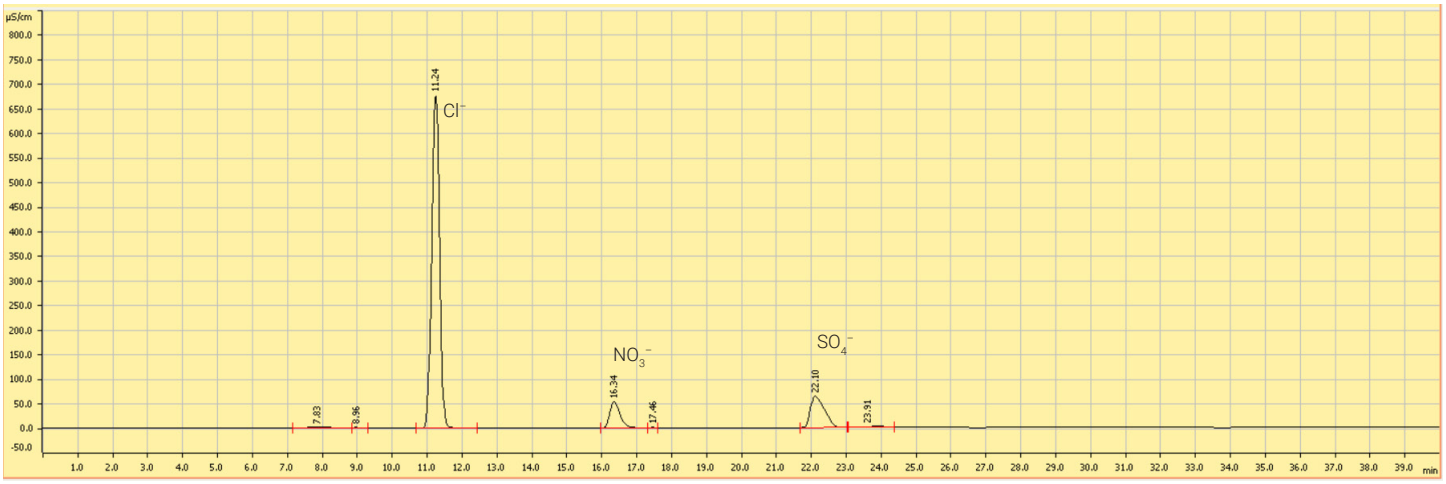

Figure 3. Conductivity chromatogram for Pennsylvania tap water.

References

1. US EPA. Drinking Water Requirements for States and Public Water Systems, Stage 1 and Stage 2 Disinfectants and Disinfection Byproducts Rules. March 9, **2022**. https://www.epa.gov/dwreginfo/stage-1-and-stage-2-disinfectants-and-disinfection-byproducts-rules. (accessed April 25, 2022).
2. US EPA. Method 552.2: Determination of Haloacetic Acids and Dalapon in Drinking Water by Liquid-Liquid Extraction, Derivatization, and Gas Chromatography with Electron Capture Detection. **1995**. https://www.o2si.com/docs/epa-method-552.2.pdf (accessed May 18, 2021).
3. US EPA. Method 557. Determination of Haloacetic Acids, Bromate, and Dalapon in Drinking Water by Ion Chromatography Electrospray Ionization Tandem Mass Spectrometry (IC-ESI-MS/MS). September **2009**. Link (accessed May 18, 2021).
4. Wu, S. *et al.* Analysis of Haloacetic Acids, Bromate, and Dalapon in Natural Waters by Ion Chromatography Tandem Mass Spectrometry. *Agilent Technologies application note* 5991-6417EN. November **2015**. https://www.agilent.com/cs/library/applications/5991-6417EN.pdf
5. Metrohm, A. G. Trace Haloacetic Acids, Dalapon, and Bromate in Water. Determination as per US EPA 557 applying IC-MS/MS. *Metrohm IC Application Note* M–15. August 2019. https://www.metrohm.com/content/metrohm/en/applications/application-notes/aa-m-001-100/an-m-015.html

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