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Food safety

Evaluation of cationic quaternary amines by ion chromatography and mass spectroscopy (IC-MS)

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Keywords

Dionex IonPac CS21-Fast-4µm column, RFIC, Reagent-Free IC, ESI-MS, single quadrupole, triple quadrupole

Goal

Demonstrate the suitability of select quaternary amines for IC-MS analysis using a Thermo Scientific™ Dionex™ IonPac™ CS21-Fast-4µm column

Introduction

Pesticide contamination in food as a potential health risk is a growing public concern, including those in the polar pesticides category, resulting in increased interest and attention by health researchers and regulatory agencies.[1] Determinations of anionic polar pesticides were previously addressed using ion chromatography with tandem mass spectrometry detection (IC-MS/MS) and single quadrupole mass spectrometry (IC-MS) methods.[2–14]

The determinations of cationic quaternary amine polar pesticides are challenging due to their similar chemistries and similar masses and mass fragments.[15–21] Previously, four quaternary amine polar pesticides (chlormequat, mepiquat, paraquat, and diquat) were analyzed by cation-exchange chromatography using the best available column technology at the time, the Thermo Scientific™ Dionex™ IonPac™ CS17 cation-exchange column. Paraquat and diquat were not chromatographically resolved and were challenging to determine by single quadrupole mass spectrometry and tandem mass spectrometry because the mass-to-charge ratios of both their parent and product ions were too similar (within 2 *m/z*) to confidently quantify. Instead, high-resolution accurate mass (HRAM) mass spectrometry detection was used to resolve these two ions.[17]

More recently, the Thermo Scientific™ Dionex™ IonPac™ CS21-Fast-4µm cation-exchange column was developed to resolve diquat, paraquat, and other analytes of interest. This was demonstrated for four quaternary amine polar pesticides, mepiquat, chlormequat, paraquat, and diquat, using IC-MS/MS[18–20] and IC-MS[21]. A recent technical note compared the peak efficiency, peak asymmetry, and resolving power of the Dionex IonPac CS21-Fast-4µm with older cation-exchange columns: Thermo Scientific™ Dionex™ IonPac™ CS17 (7 µm), CS19-4µm, and CS20 (5 µm) columns for a larger group of quaternary amines.[22] This technical note focused on biologically active quaternary amine compounds (choline, carbachol, bethanechol, acetylcholine, methacholine, and succinylcholine) along with the quaternary amine pesticides (chlormequat, mepiquat, paraquat, and diquat) and used suppressed conductivity detection. The conclusion was that the analyte peaks had the best symmetries and highest efficiencies when using the Dionex IonPac CS17 and CS21-Fast-4µm columns.

However, only four cationic quaternary amine polar pesticides were evaluated on the Dionex IonPac CS21-Fast-4µm cation-exchange column using IC-MS. In this technical note, fifteen additional analytes of interest are evaluated using the Dionex IonPac CS21-Fast-4µm cation-exchange column with an electrolytically generated methanesulfonic acid (MSA) gradient and detected by suppressed conductivity detection and mass spectrometry using a Thermo Scientific™ ISQ™ EC single quadrupole mass spectrometer. This technical note provides the IC-MS conditions for twelve of the fifteen analytes. Three analytes, 1,2,4-triazole, difenzoquat, and propamocarb, are unsuitable using the Dionex IonPac CS21-Fast-4µm column for IC-MS analysis.

Experimental

Equipment

- Thermo Scientific™ Dionex™ ICS-6000 HPIC™ system*
  - Dual Pump DP module, isocratic configuration (P/N 22181-6009)* with pump_1 to deliver DI water for eluent generation and pump_2 to deliver DI water as suppressor regenerant.
  - Eluent Generator EG module (P/N 22181-60019)
  - Detector Chromatography DC module with two 6-port injection valves (P/N 22181-600499)**
  - CD Conductivity Detector (P/N 079829)
- Thermo Scientific™ Dionex™ AS-AP Autosampler with temperature control option (P/N 074926)
  - 100 µL syringe (syringe P/N 074305)
  - 1 to 3 8-position sample trays for 10 mL vials (P/N 069877) typically used for standards
  - 1 to 3 19-position sample trays for 10 mL vials (P/N 074938)
- Thermo Scientific™ ISQ EC™ single quadrupole mass spectrometer with HESI-II probe (P/N ISQEC-IC)

*Alternative IC pump configuration: Thermo Scientific ICS-6000 with single pump SP module (P/N 22181-6003) or Thermo Scientific™ Dionex™ Integrion™ HPIC™ system (P/N 22153-60305). The Thermo Scientific™ Dionex™ Auxiliary Pump AXP-MS (P/N 063973) is used to deliver DI water for the suppressor regenerant.

**Alternative valve configuration: Thermo Scientific ICS-6000 DC with single injection valve (P/N 22181-60047) and Automation Manager with one (IC PEEK) 6-port valve (P/N 075952) used as the diverter valve or a Dionex Integrion system with a second 6-port injection valve (P/N 22153-62027).

Consumables

Table 1 lists the consumable products needed for the IC-MS system.

Table 1. Consumables list for the Dionex ICS-6000 HPIC system coupled to the ISQ EC single quadrupole mass spectrometer

Product name	Description	P/N
Thermo Scientific™ Dionex™ IC PEEK Viper™ fitting tubing assembly kit	Dionex IC Viper fitting assembly kit for the Dionex ICS-6000 HPIC dual microbore system with CD	302965
Thermo Scientific™ Dionex™ EGC 500 MSA Eluent Generator cartridge	Cation eluent generator cartridge for HPIC high pressure systems	075779
Thermo Scientific™ Dionex™ CR-CTC™ III Electrolytic trap column*	Continuously regenerated cation trap column required for determining quaternary amine analytes, as in this technical note. The cable is standard for the Dionex Integrion HPIC and ICS-6000 HPIC instruments.	104-30001
Thermo Scientific™ Dionex™ ICS-6000 EG Eluent Generator kit	Degasser installed after Dionex CR-CTC III trap column and before the injection valve, used with eluent generation	075522
Thermo Scientific™ Dionex™ CDRS™ 600 suppressor, 2 mm	Suppressor for 2 mm cation columns	088670
Thermo Scientific™ Dionex™ IonPac™ CG21-Fast-4µm guard column	Cation guard column, 2 × 30 mm	303349
Thermo Scientific™ Dionex™ IonPac™ CS21-Fast-4µm analytical column	Cation analytical column, 2 × 150 mm	303348
Thermo Scientific™ Dionex™ AS-AP autosampler vial kit	10 mL polystyrene, package of 100 caps and septa	055058
Thermo Scientific™ Dionex™ IC PEEK Viper™ sample loop	10 µL Dionex IC PEEK Viper sample loop	302895
Thermo Scientific™ Dionex™ IC PEEK Viper tubing, 0.007" i.d. (0.01778 cm) × 34" length (864 mm)	Dionex IC PEEK Viper tubing connection from IC to mass spectrometer	088916
IC-MS Installation Kit	IC-MS installation kit includes tubing, mixing tee	22153-62049

*P/N for cable adapter to Dionex ICS-5000+ HPIC system: 22181-98150

Reagents

- Degassed deionized (DI) water, ASTM Type I grade[24]
- Fisher Scientific™ reagents
  - Chlormequat (2-chloroethyl(trimethyl)azanium), 98%, P/N AC215421vv000 (CAS 999-81-5)
  - Cyromazine (2-*N*-cyclopropyl-1,3,5-triazine-2,4,6-triamine), P/N AC467530250 (CAS 66215-27-8)
  - PESTANAL™ Propamocarb, MilliporeSigma™ Supelco™, (propyl *N*-[3-(dimethylamino)propyl]carbamate, P/N 11-101-3681 (CAS 24579-73-5)
  - 1,2,4-Triazole, 99%, P/N AAA1159736 (CAS 288-88-0)
  - Trimethylsulfonium 98% (trimethylsulfonium methyl sulfate, P/N AC386610250 (CAS 676-84-6)
- Thermo Scientific™ reagents
  - Diethanolamine, (2-(2-hydroxyethylamino)ethanol), 99%, P/N AAA1338936 (CAS 111-42-2)
  - 1,1-Dimethylhydrazine, P/N S1627 (CAS 57-14-7, SPEX CertiPrep™)
  - Diquat dibromide, monohydrate (1,1'-Ethylene-2,2'-dipyridylium dibromide), SPEX CertiPrep™, P/N S1752 (CAS 6385-62-2)
  - Melamine (1,3,5-triazine-2,4,6-triamine), 99%, P/N AC125350100 (CAS 108-78-1)
  - Mepiquat chloride (1,1-Dimethylpiperidinium chloride), 98%, Honeywell Fluka™ PESTANAL™, P/N 11-101-3665 (CAS 24307-26-4)
  - Paraquat dichloride, tetrahydrate (1-methyl-4-(1-methylpyridin-1-ium-4-yl) pyridin-1-ium;dichloride), SPEX CertiPrep, P/N S2915 (CAS 1910-42-5)
  - Streptomycin sulfate (2-[(1R,2R,3S,4R,5R,6S)-3-(diaminomethylideneamino)-4-[(2R,3R,4R,5S)-3-[(2S,3S,4S,5R,6S)-4,5-dihydroxy-6-(hydroxymethyl)-3-(methylamino)oxan-2-yl]oxy-4-formyl-4-hydroxy-5-methyloxolan-2-yl]oxy-2,5,6-trihydroxycyclohexyl]guanidine), P/N AC455341000 (CAS 3810-74-0)
  - Triethanolamine, 98%, P/N AAL0448636 (CAS 102-71-6)
- Other
  - Difenzoquat, methyl sulfate, ChemService™ P/N N-11721-250MG (CAS 43222-48-6)
  - Nereistoxin oxalate (*N,N*-dimethyldithiolan-4-amine, FUJIFILM™ Wako Pure Chemical™ U.S.A Corporation P/N 141-04161 (CAS 1631-58-9)

Software

Thermo Scientific™ Chromeleon™ Chromatography Data System (CDS) 7.2 version 10 with MS driver or 7.3 version (MS driver is included)

IC-MS conditions

Method 1

Columns	Dionex IonPac CG21-Fast-4µm guard (2 × 30 mm) and Dionex IonPac CS21-Fast-4µm analytical column (2 × 150 mm)		
MSA gradient	1 mM MSA (0 to 0.1 min), 1–3.2 mM (0.1 to 4 min), 3.2–15 mM (4 to 10 min), 15–40 mM (10 to 14 min), 40 mM (14 to 17 min), 1 mM (17.1 to 20 min)		
Eluent source	Dionex EGC 500 MSA eluent cartridge, Dionex CR-CTC III trap column and HP EG degas kit		
Flow rate	0.30 mL/min		
Injection volume	10 µL		
Column temperature	40 °C		
Detection/Suppressor compartment	20 °C		
Detection 1	Suppressed conductivity, Dionex CDRS 600 suppressor, 2 mm, 47 mA, constant current, and external water modes		
Suppressor regenerant	DI water by DP Pump 2 at 0.3 mL/min		
Conductivity	Background: <1 µS/cm, Noise: <1 nS/cm		
IC-MS system backpressure	~3,000 psi (100 psi = 689.5 kPa)		
DC Valve 2 functioning as a diverter valve	Timing (min)	Valve position	IC flow path
	Prerun	DC.InjectValve_2.InjectPosition	Divert IC flow away from MS. DI water for Suppressor Regen flows through diverter valve to MS
	0.0 Run	Same	Start IC acquisition
	2.0 min	DC.InjectValve_2.LoadPosition	IC flow to MS Start MS acquisition
	20 min	DC.InjectValve_2.InjectPosition	Divert away from MS
Detection 2 (ISQ-EC)	ISQ EC single quadrupole mass spectrometer, HESI-II, +ESI, +3,500 V, Full Scan 50–600 *m/z*, SIM mode		
Flow (N_2)	Sheath: 35 (psi), Aux: 4 (psi), Sweep: 1 (psi)		
MS temperatures	Vaporizer: 300 °C, Ion transfer tube: 350 °C		
Desolvation solution	None		
SIM conditions	Method type:		Component mode, Advanced
	Polarity:		Positive
	Scan time (s):		0.5 s
	Use calibrated RF lens:		(check box)
IC-MS run time	20 min		

Method 2, Nereistoxin

MSA gradient	1 mM MSA (0 to 0.1 min), 1–3.2 mM (0.1 to 4 min), 3.2–15 mM (4 to 10 min), 15–40 mM (10 to 14 min), 40 mM (14 to 20 min), 1 mM (20.1 to 30 min)
Detection 2	ISQ EC single quadrupole mass spectrometer
MS conditions	Same as Method 1
SIM	*m/z* CID: 10 V
IC-MS run time	30 min
Desolvation solution	None
IC-MS run time	30 min

Method 3, Cyromazine

MSA gradient	1 mM MSA (0 to 0.1 min), 1–3.2 mM (0.1 to 4 min), 3.2–15 mM (4 to 10 min), 15–75 mM (10 to 20 min), 1 mM (20 to 30 min)
Detection 1	Suppressed conductivity, same as Method 1, 88 mA
Detection 2 (ISQ-EC)	Thermo Scientific ISQ EC single quadrupole mass spectrometer, same conditions as Method 1
SIM	*m/z* 176, CID: 10 V

Method 4, Propamocarb

MSA gradient	30 mM MSA (0 to 0.1 min), 30–85 mM (0.1 to 4 min), 85 mM (4 to 95 min), 85–30 mM (95 to 95.5 min), 30 mM (95.5 to 100 min)
Detection 1	Suppressed conductivity, Dionex CDRS 600 suppressor, 2 mm, 80 mA, constant current and external water modes
Suppressor regenerant:	DI water by DP Pump 2 at 0.3 mL/min.
Detection 2 (ISQ-EC)	ISQ EC single quadrupole mass spectrometer, same as Method 1
SIM	*m/z* 189, CID: 10 V

Instrument setup and installation

Physical and electronic configuration

The IC-MS configuration requires two pumps and two IC PEEK injection valves. The Dionex ICS-6000 system is a modular, high pressure IC system. For this technical note, a Reagent-Free™ IC (RFIC™) dual ICS-6000 system is used. The separation uses one pump of the dual system: System 1 (Figure 1). The second pump of the DP module delivers DI water as the regenerant solution to the suppressor. The second DC Injection Valve 2 is used as the IC PEEK diverter valve to facilitate the flow to the suppressor regenerant (Inject mode) and to the MS (Load mode) (Figure 2).

In IC-MS applications, a PEEK valve should be used as the diverter valve (Figure 2). The valve timing is intended to divert most of the sample matrix away from the mass spectrometer (For the applications in this technical note, 0 to 2 min and >30 min). In IC-MS, a heated electrospray source transfers chromatographically separated ions in solution to ions in a gas stream. A make-up solvent is not needed.

For the best results, position the Dionex ICS-6000 system near the MS source. Install the power and USB cables, and power up the IC, autosampler, and computer. Add DI water to the eluent bottles and prime the pumps. Use Dionex IC PEEK Viper fittings (from the IC-MS installation kit) to complete the connections from the IC effluent to the diverter valve (Inj.Valve_2) to the MS source.

Install the mass spectrometer according to the operating manual.[25]

Electronic configuration

Chromeleon CDS was used exclusively for the configuration and operation. To electronically configure the IC system, start the Thermo Scientific™ Chromeleon™ Instrument Services program,

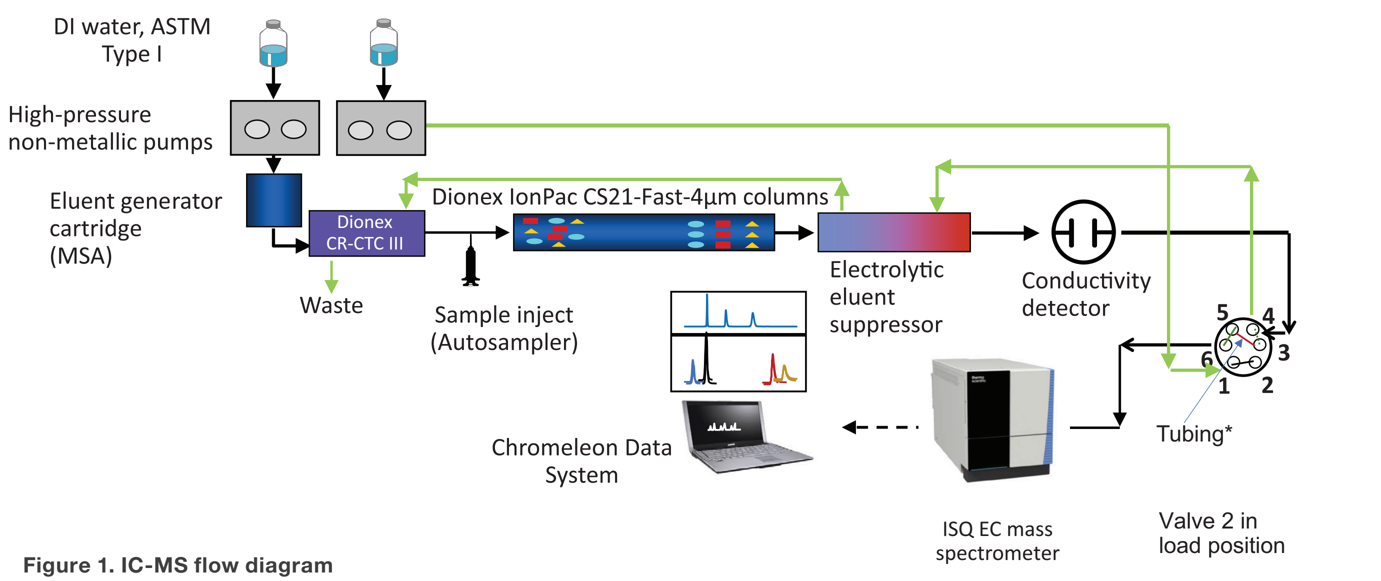


Figure 1. IC-MS flow diagram

* 3-5 in (7.6-12 cm) long red PEEK tubing (0.005 in i.d.; 0.013 cm i.d.)

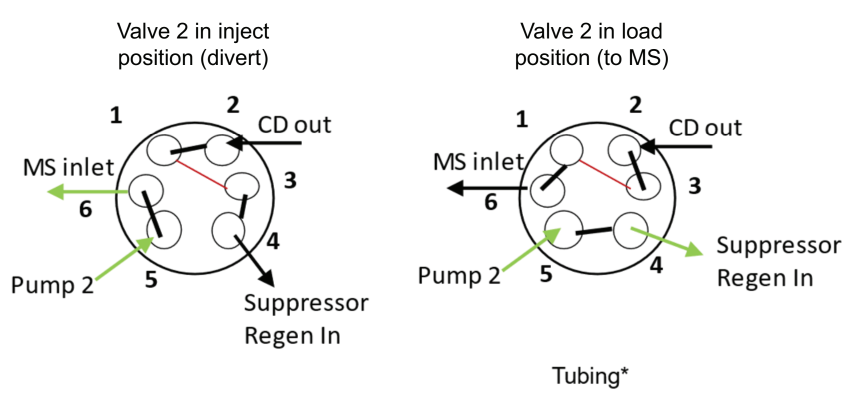


Figure 2. Valve 2 (diverter) positions

then start the Instrument Controller program by selecting the *Configure Instruments* link. Add the ICS-6000 system modules: DP, EG, and DC, the AS-AP Autosampler, and the mass spectrometer as described in Table 2. Check the configuration for any errors and correct as needed. Save and close the configuration.

Plumbing the Dionex ICS-6000 HPIC system

Plumb the Dionex ICS-6000 IC system as a standard Reagent-Free IC (RFIC) system as shown in Figure 1. Use the IC PEEK Viper fittings as indicated on their labels. Install a 2,000 psi backpressure tubing loop on Injection Valve 2 position 6 for DP.Pump_2 to maintain similar pressure as the IC system and to maintain backpressure well above the 500 psi lower limit. Temporarily direct the liquid flow away from the ISQ EC mass spectrometer until the IC system and consumables are fully conditioned. The schematics are also illustrated on the inside doors of the Dionex ICS-6000 IC system. Further information can be found in the operator's and installation manuals.[26,27] Direct the waste lines to waste containers.

Conditioning consumable devices

Important: If consumable tracking is installed, do not remove consumable tracking tags on the columns and consumable devices. These tags are required for consumables monitoring functionality.

Temporarily direct the liquid flow away from the mass spectrometer until the IC system and consumables are fully conditioned. Hydrate and condition the Dionex EGC 500 MSA eluent generator cartridge and Dionex CR-CTC III continuously regenerated trap column according to product manuals or the instructions in the drop-down menu (Chromeleon Console, under Consumables).[28,29] The Dionex CR-CTC III trap column is required for this application. Note: Long-term use of a Dionex CR-CTC II or Dionex CR-CTC 500 trap column for this application will damage the Dionex IonPac CS21-Fast-4µm column.

Table 2. Electronic configuration parameters

Module	Tab	Action
ICS-6000 DP	General	Select *Browse*, select the serial number to link module to Instrument
	Device	Link Pump_1 and Pump_2 to Instrument
	Top Pump	Set minimum pressure to *500* psi
	Bottom Pump	Set minimum pressure to *500* psi
	Error Levels	Set "System pressure below limit" to *Abort*
ICS-6000 EG	General	Select serial number to link module to instrument
	Cartridges	Link to instrument. Check *EGC_1* and link to *Pump_1*
ICS-6000 DC	General	Select the serial number to link the module to the instrument, select instrument
	Detectors	Double click on CD, Link to *Pump_1*
	Thermo Controls	Check *Compartment_TC* and Column_TC
	Suppressors	Double click *Suppressor_1*, Link to *Pump_1*
	High Pressure Valves	Double click *InjectValve_1*, Link to *Pump_1*, select control by autosampler
		Double click I*njectValve_2*, *Link to Pump_1*, select control by DC
AS-AP autosampler	General	Select serial number to link module to instrument
	Sharing	If this option is present, select instrument
	Segments/Pump Link	Select *10 mL PolyVials* for "Red", "Blue", and Green. Leave the pump and TTL links empty.
	Options	Select *1200 buffer loop, 250 µL syringe, Temperature control,* and *Push mode*. Enter "*10*" µL in sample loop.
ISQ mass spectrometer	Add module	Mass spectrometry, *ISQ ICMS family*
	General	Device model: *ISQ EC*
		Deselect the hardware inject synchronization. This is not needed for Chromeleon CDS.
		Remote Start: Select *ActiveLow* (ASAP autosampler)
		Deselect *Split flow, Fraction collection, Warn on source change and Simulation mode* boxes.
		Vacuum pressure units: *Torr*
		Source gas pressure units: *psig*
	Maintenance	Enable all boxes. Select "OK".
	Associate Pump Flow	Optional: Select *Associate* — a pump box will automatically enter flow rate when setting source conditions. If using this option, select pump_1 used for eluent flow.

Condition the columns as described in the Dionex IonPac CS21-Fast-4µm product manual[29] or the Consumables Conditioning instructions in the drop-down menu (Chromeleon Console, under Consumables) using 4 mM MSA, 40 °C at 0.30 mL/min for 30 min or more while directing the effluent to waste. Install the conditioned columns according to Figure 2.

To hydrate the Dionex CDRS 600 suppressor, follow the instructions in the Suppressor Installation Checklist included with the suppressor.[30,31]

Install the suppressor according to Figure 1 and ensure that the suppressor is within backpressure specifications. For cation suppressors, optimum results are achieved by minimizing the hydrating and waiting times to those stated in the suppressor installation checklist, immediately installing the suppressor, starting the pump, eluent generation and CR-CTC III trap column, and powering the suppressor.

Plumbing the IC system to the MS system

Use Dionex IC PEEK Viper fittings (P/N 088916) to complete the connections from the IC system effluent to the diverter valve (Inj.Valve_2) to the MS source. Monitor and minimize backpressure to the suppressor (<100 psi) to prevent damage to the suppressor. Check the backpressure to the suppressor as part of the start-up and daily checklist.

Additional maintenance of the Dionex AS-AP autosampler may be required after six months due to cross-contamination and resulting carryover of diquat and paraquat. The following parts should be replaced every six months: AS-AP injection port, transfer line tubing, and buffer loop.

Results and discussion

The chemical structures of the compounds evaluated for separation on the Dionex IonPac CS21-Fast-4µm column are shown in Table 3.[32]

Table 3. Analytes of interest

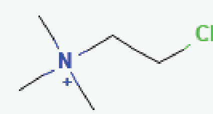
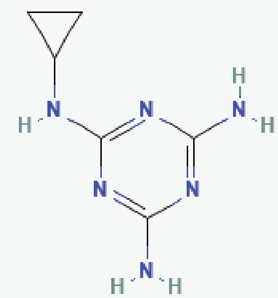
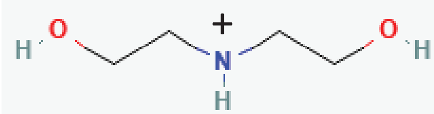
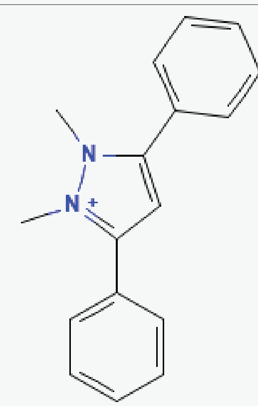
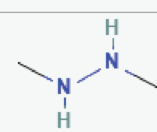
Analyte	Molecular weight (g/mol)	Chemical formula	Chemical structure
Chlormequat	122.62	$C_5H_{13}ClN^+$	
Cyromazine	166.18	$C_6H_{10}N_6$	
Diethanolamine	105.14	$(CH_2CH_2OH)_2NH$	
Difenzoquat	249.33	$C_{17}H_{17}N_2^+$	
Dimethylhydrazine	60.10	$H_2NN(CH_3)$	

Table 3. Analytes of interest (continued)

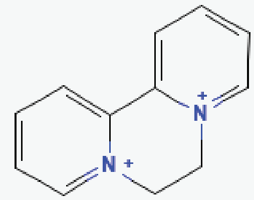
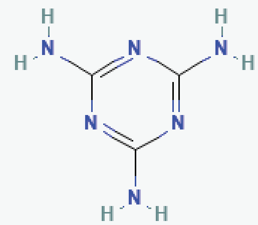
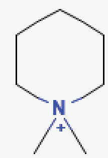
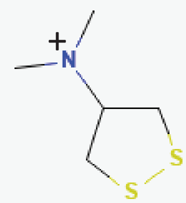
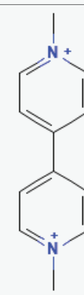
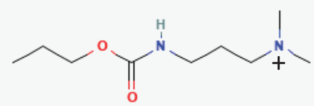
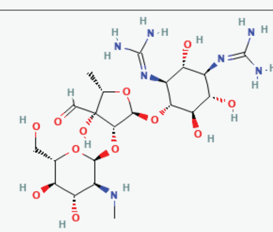
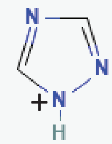
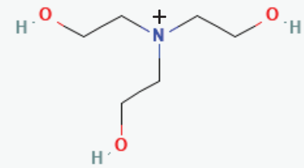
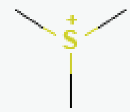
Analyte	Molecular weight (g/mol)	Chemical formula	Chemical structure
Diquat	184.24	$C_{12}H_{12}N_2^{+2}$	
Melamine	126.12	$C_3N_3(NH_2)_3$	
Mepiquat	114.21	$C_7H_{16}N^+$	
Nereistoxin	149.3	$C_5H_{11}NS_2$	
Paraquat	186.25	$C_{12}H_{14}N_2^{+2}$	
Propamocarb	188.27	$C_9H_{20}N_2O_2$	
Streptomycin 57-92-1	581.6	$C_{21}H_{39}N_7O_{12}$	

Table 3. Analytes of interest (continued)

Analyte	Molecular weight (g/mol)	Chemical formula	Chemical structure
1,2,4-Triazole	69.07	$C_2H_3N_3$	
Triethanolamine	149.19	$(CH_2OHCH_2)_3N$	
Trimethylsulfonium	77.17	$C_3H_9S^+$	

*PubChem[32]

The analytes were evaluated using Method 1 as the starting conditions to screen for their suppressed conductivity and mass spectrometry responses. Figures 3A, 3B, and 4–7 show the IC and SIM chromatograms of diethanolamine, triethanolamine, trimethylsulfonium, chlormequat, mepiquat, paraquat, and diquat. The analytes eluted within 16 min with good responses by suppressed conductivity and mass spectrometry detections. However, the other six analytes (dimethylhydrazine, melamine, nereistoxin, cyromazine, propamocarb, difenzoquat, and 1,2,4-triazole) required further experiments (Figures 8–11). Dimethylhydrazine had a strong response by suppressed conductivity but a poor response by mass spectrometry. Nereistoxin, melamine, and cyromazine had poor responses by suppressed conductivity but strong mass spectrometry responses. Propamocarb was retained on the Dionex IonPac CS21-Fast-4µm column for more than 80 min at 85 mM MSA. Difenzoquat and 1,2,4 triazole were detected neither by suppressed conductivity nor mass spectrometry. Table 4 summarizes these results.

Chlormequat, mepiquat, paraquat, diquat, diethanolamine, triethanolamine, and trimethylsulfonium

Figures 3A, 3B, and 4–7 show that these six analytes have good responses by suppressed conductivity and mass spectrometry detections. Figures 3A and 3B show the IC and SIM chromatograms of chlormequat, mepiquat, paraquat, and diquat using Method 1.

Table 4. Summary

Analyte	Method	IC retention time (min)	Elution concentration (mM MSA)	MS retention time (min)	Scan window (min)	*m/z*	CID (V)	Comment
Dimethyl hydrazine	1	2.8, 3.0	7.6, 7.7					MS response is poor
Diethanolamine	1	5.4	6.3	5.5	4–7	106	10	
Triethanolamine	1	5.7	6.4	5.75	4–7	150	10	
Trimethylsulfonium	1	6.5	3.3	6.8	5–8	77	10	
Melamine	1	7.85	11	7.8	6–9	127	10	Poor IC chromatogram
Chlormequat	1	7.4	9.6	8.0	7–10	122	10	
Mepiquat	1	8.3	8.2	9.1	7–10	114	10	
Nereistoxin	2	15	40	15.5	14–25	150	10	Poor IC chromatogram
Streptomycin	1	15.10	40	15.16	14–16	600	5	
Paraquat	1	15.7	40	16.5	15–18	183	10	
Diquat	1	16.2	40	17.1	16–19	185	10	
Cyromazine	3	16.4	40	17.6	16–20	70	10	Poor IC chromatogram
Propamocarb	4	82	85	82.03	70–100	189	10	
Difenzoquat	1	ND*		ND*		250		
1,2,4-Triazole	1	ND*		ND*		70		

*ND = not detected by suppressed conductivity or mass spectrometry

Method 1

Columns:	Dionex IonPac CG21-Fast-4µm guard, Dionex IonPac CS21-Fast-4µm analytical, 2 mm i.d.
Eluent source:	Dionex EGC-500 MSA cartridge, Dionex CR-CTC III trap column, Dionex high pressure degasser
MSA gradient:	1 mM MSA (0 to 0.1 min), 1–3.2 mM (0.1 to 4 min), 3.2–15 mM (4 to 10 min), 15–40 mM (10 to 14 min), 40 mM (14 to 20 min), 1 mM (20.1 to 30 min)
Flow rate:	0.38 mL/min
Injection vol.:	10 µL
Column temp.:	30 °C
Detector oven temp.:	20 °C
Detection:	Suppressed conductivity, Dionex CDRS 600, 2 mm, external water mode by pump 2 @ 0.3 mL/min, 57 mA
System pressure:	2,800 psi
Run time:	20 min

Peaks:	mg/L		mg/L
1. Potassium	2	5. Calcium	5
2. Mepiquat	5	6. Paraquat	5
3. Chlormequat	5	7. Diquat	5
4. Magnesium	1		

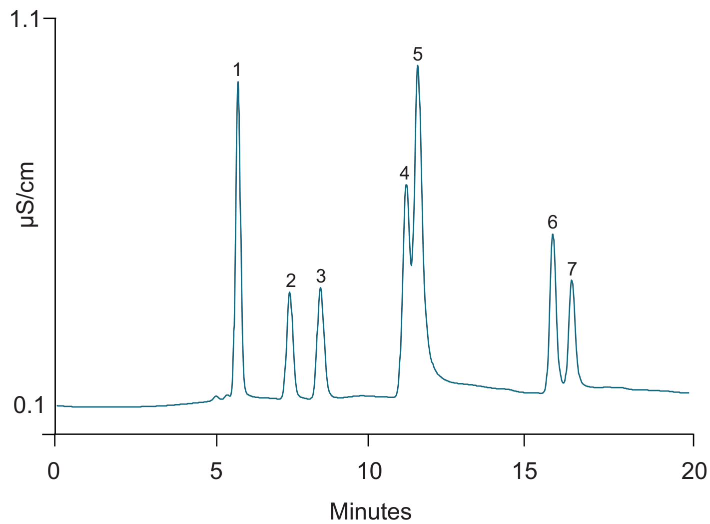


Figure 3A. IC chromatogram of mepiquat, chlormequat, paraquat, and diquat with three inorganic cations

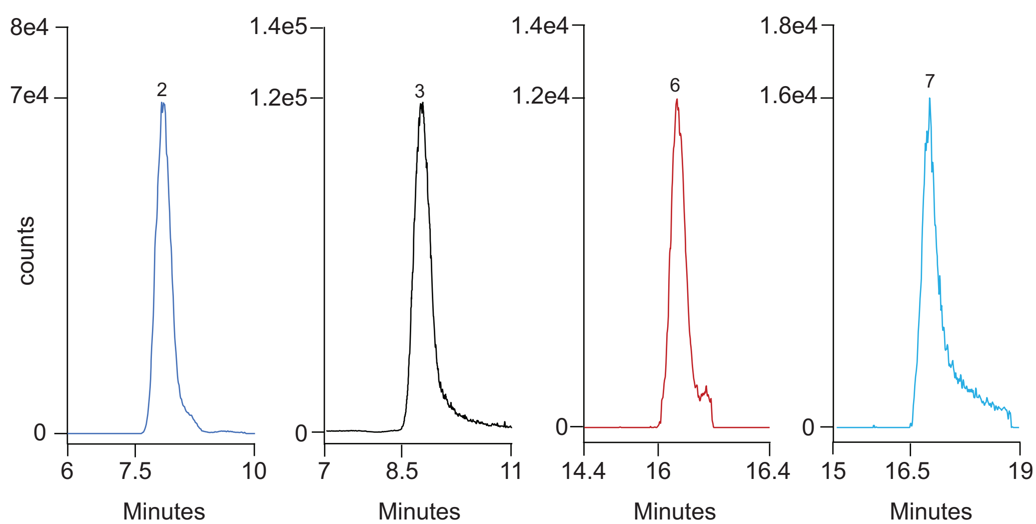


Figure 3B. SIM chromatogram of mepiquat, chlormequat, paraquat, and diquat. SIM conditions: +3,000 V, N2 gas (psi): sheath gas (50), aux gas (4), and sweep gas (0.2), Temp. (°C): vaporization (250), ion transfer (250), CID (V): 10

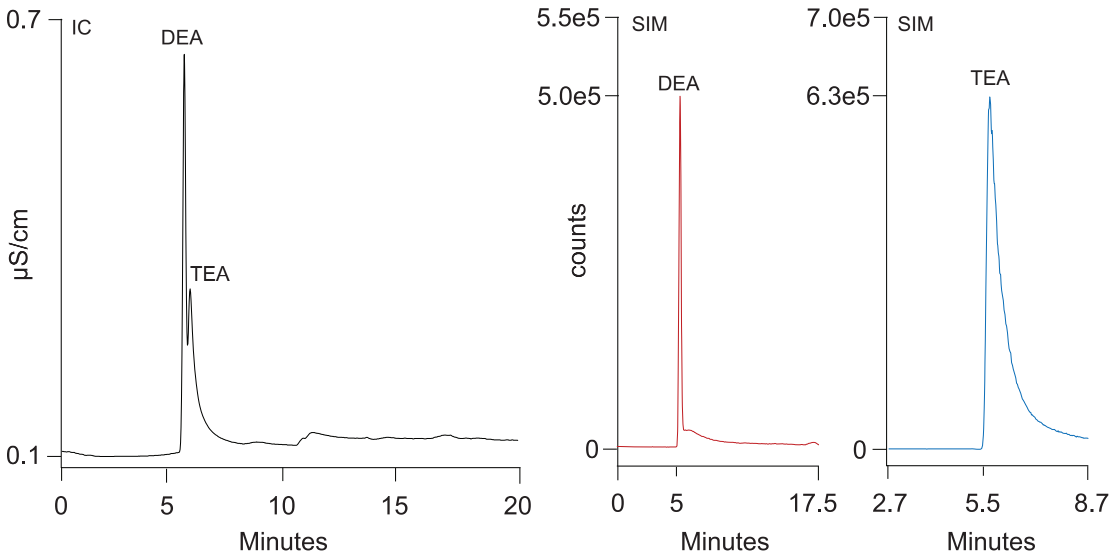


Figure 4. IC and SIM chromatograms of 10 mg/L diethanolamine (DEA) and 10 mg/L triethanolamine (TEA) (Method 1)

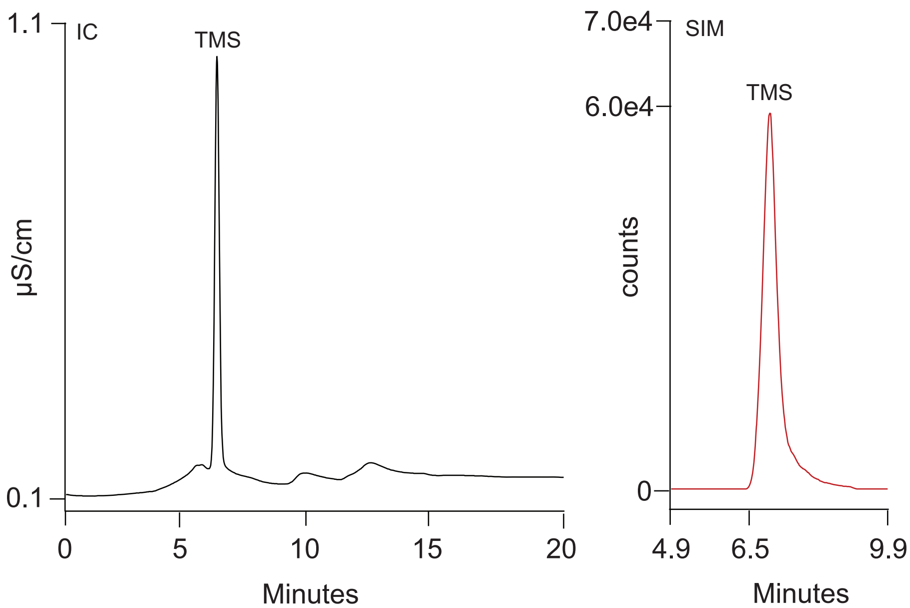


Figure 5. IC and SIM chromatograms of 10 mg/L trimethylsulfonium (TMS) (Method 1)

Figure 4 shows the IC and SIM chromatograms of 10 mg/L DEA and TEA using Method 1. Both DEA and TEA show good responses by IC and mass spectrometry detections.

Figures 5 and 6 show the chromatograms of 10 mg/L TMS and 40 mg/L streptomycin. The collision induced dissociation (CID) voltage was reduced from 10 V (Method 1) to 5 V for improved detection of streptomycin.

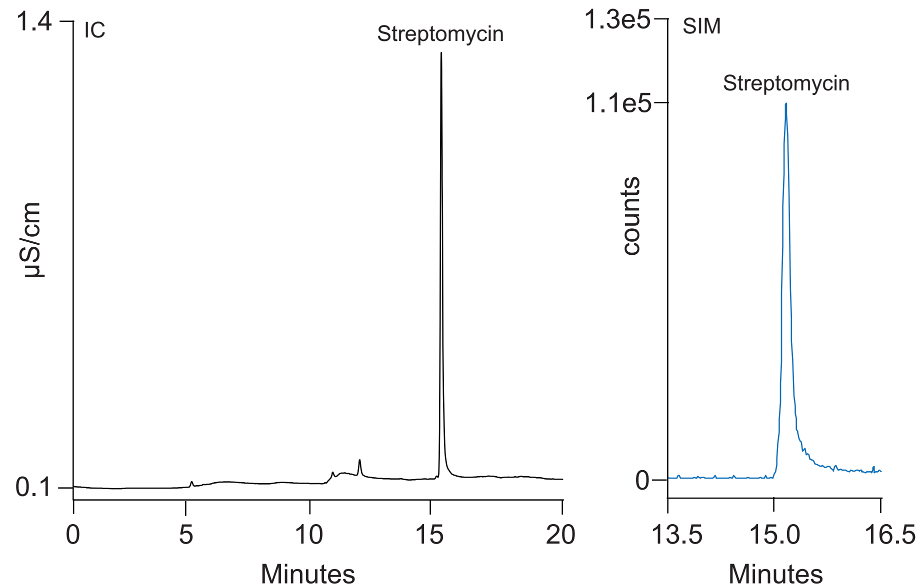


Figure 6. IC and SIM chromatograms of 40 mg/L streptomycin (Method 1, CID: 5 V)

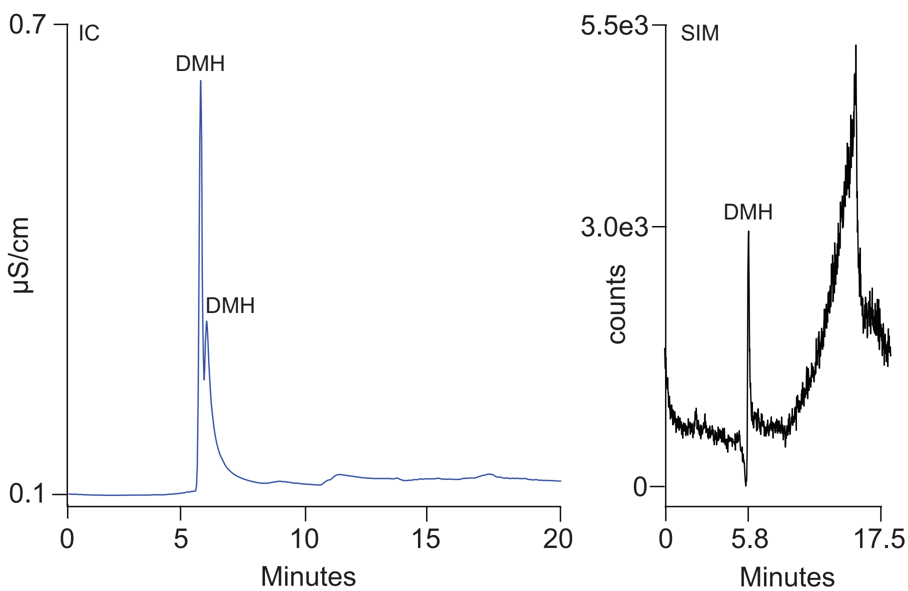


Figure 7. IC and SIM chromatograms of 10 mg/L dimethylhydrazine (Method 1)

Dimethylhydrazine

Figure 7 shows the IC and SIM chromatograms of 10 mg/L dimethylhydrazine (DMH). To try to improve the mass spectrometer's response, the CID voltage was applied at 0, 5, and 10 V with negligible effect. Additionally, DMH was also evaluated using mass spectrometry conditions applied for an earlier amine application.[23] The ionization voltage (3,000 V) and ion transfer (250 °C) and vaporization (250 °C) temperatures were lower than Method 1 conditions. The sheath gas flow conditions (50 psi) were higher than those in Method 1. CID voltage was applied at the three levels. The experimental conditions did not improve the SIM response, subsequently Figure 7 shows Method 1 results. DMH is suitable for IC-MS determinations, although suppressed conductivity detection should be used for quantitation and the SIM results used for confirmation.

Nereistoxin, melamine, cyromazine, propamocarb, difenzoquat, and 1,2,4-triazole

Figures 8–11 show IC and SIM chromatograms of nereistoxin, melamine, and cyromazine. These analytes had poor responses by suppressed conductivity but strong responses by mass spectrometry. To improve the suppressed conductivity responses and make identification easier, the standard concentrations were increased from 10 mg/L to 100 mg/L. This strategy was effective for nereistoxin and melamine. SIM responses were strong for a 10 to 40 mg/L standard, with responses in the 10^5 range.

Figure 8 shows a broad IC peak of 100 mg/L nereistoxin eluting near the expected retention time (Method 2). In Figure 9, a tiny peak was detected in the expected region for melamine. The IC chromatogram of 100 mg/L cyromazine using Method 1 was the same as the DI water blank, that is, no IC response was detected. To improve the suppressed conductivity response of cyromazine, an eluent gradient was applied at 10 min from 15 to 75 mM MSA (Method 3). Figure 10 shows that there was no improvement; the IC chromatogram was the same as the DI water blank. These experiments show that IC-MS is a suitable method for nereistoxin, melamine, and cyromazine, despite the poor suppressed conductivity detection responses. Detection and quantitation must be done by mass spectrometry.

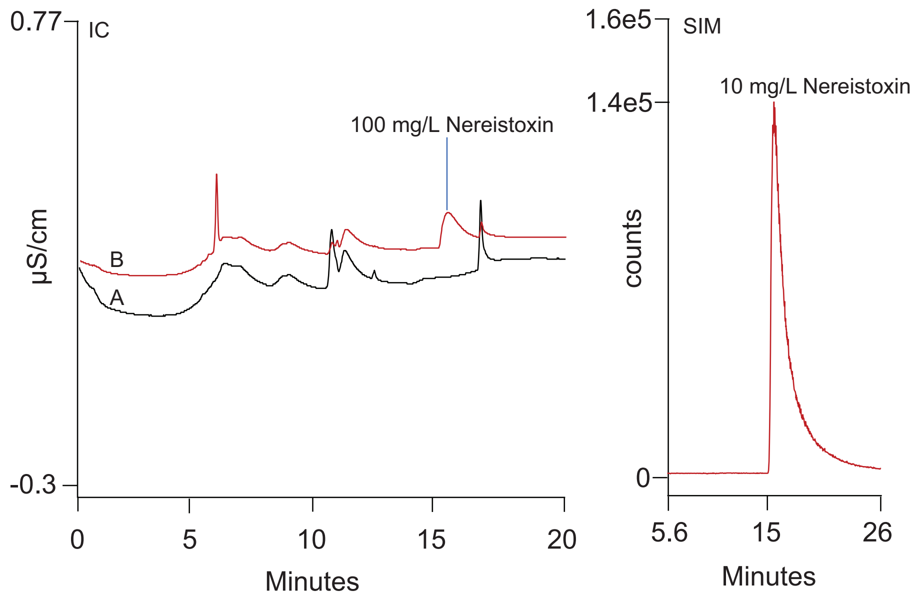


Figure 8. IC and SIM chromatograms of (A) DI water and (B) nereistoxin (Method 2)

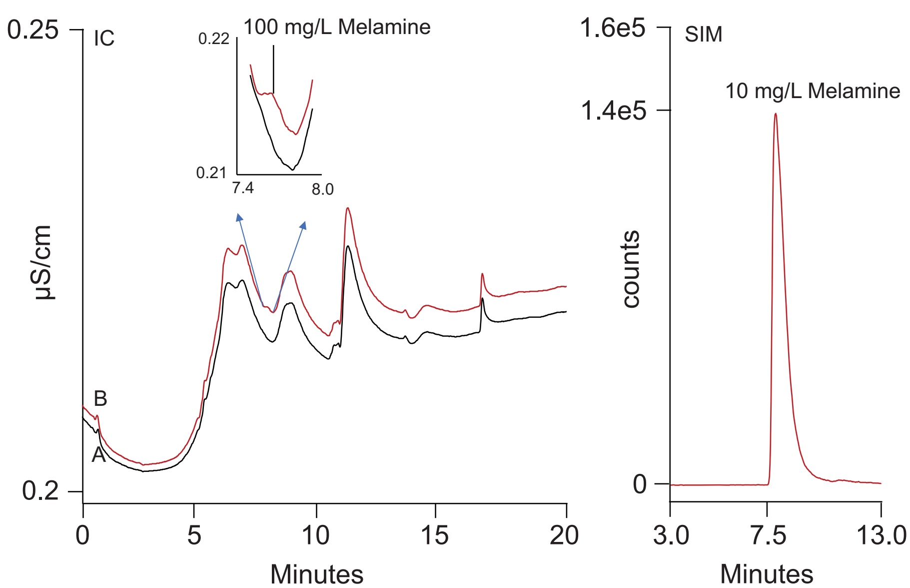


Figure 9. IC and SIM chromatograms of (A) DI water and (B) melamine (Method 1)

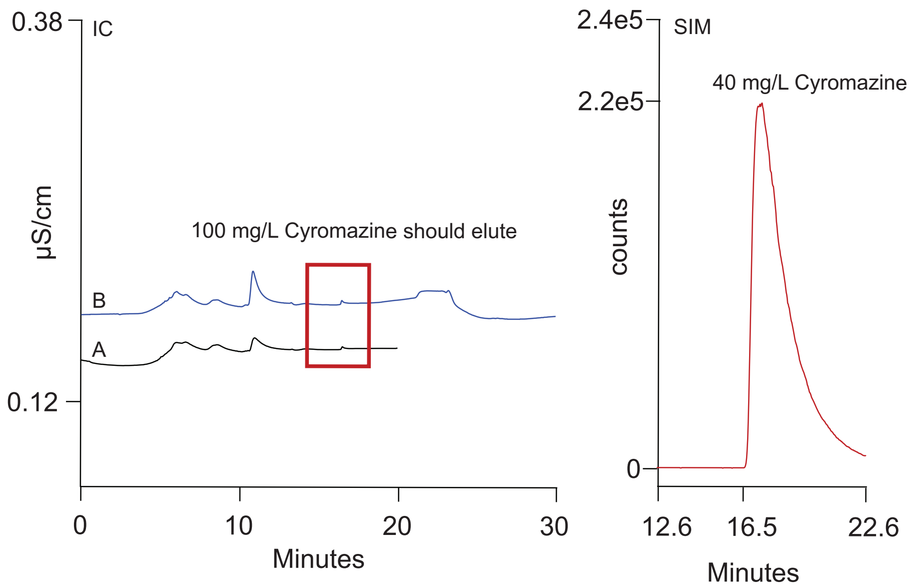


Figure 10. IC and SIM chromatograms of (A) DI water and (B) cyromazine (Method 3)

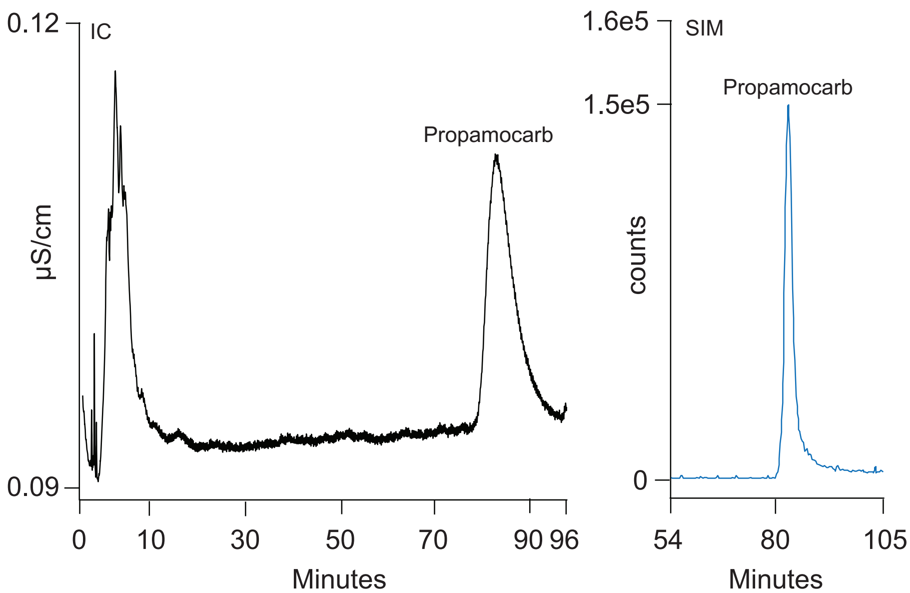


Figure 11. IC and SIM chromatograms of 10 mg/L propamocarb (Method 4)

Figure 11 shows the IC and SIM chromatograms of propamocarb. Propamocarb is highly retained on the Dionex IonPac CS21-Fast-4µm column, eluting at 80 min despite using a gradient starting at 30 mM MSA and holding at 85 mM MSA for 95 min (Method 4). Propamocarb has good responses by both detectors, however the long retention time makes it impractical for IC-MS.

Difenzoquat and 1,2,4-triazole were detected by neither suppressed conductivity nor mass spectrometry using the evaluated methods (not shown).

Conclusion

The feasibility of using the Dionex IonPac CS21-Fast-4µm column and IC-MS was determined for fifteen analytes, including the four cationic polar pesticides (mepiquat, chlormequat, paraquat, and diquat) previously evaluated.[21] Twelve analytes were found suitable for IC-MS methods. Chlormequat, mepiquat, paraquat, diquat, diethanolamine, triethanolamine, and trimethylsulfonium had strong responses by suppressed conductivity and mass spectrometry. DMH had a strong response by suppressed conductivity, but the mass spectrometry response was inadequate. DMH is suitable for IC-MS by quantitating on the suppressed conductivity channel and confirming on the mass spectrometry channel. Determinations of nereistoxin, melamine, and cyromazine must be done on the mass spectrometry channel because their suppressed conductivity responses were negligible.

Three analytes—propamocarb, difenzoquat, and 1,2,4-triazole—were found to be unsuitable for IC-MS. Propamocarb is impractical due to its 80 min retention time. Difenzoquat and 1,2,4-triazole were not detected by either detector. More information on similar application documents can be found on the Thermo Scientific Digital Library website.[34]

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