PRODUCT MANUAL

IONPAC® AG12A GUARD COLUMN

(4 x 50 mm, P/N 079801)
(2 x 50 mm, P/N 046056)

IONPAC® AS12A ANALYTICAL COLUMN

(4 x 200 mm, P/N 046034)
(2 x 200 mm, P/N 046055)

QUICKSTART STEPS AND LINKS
Click blue text below to get started.

1. See Section 4, “Operation”. Note operation precautions and chemical purity requirements.
2. See Section 5.1, “Preparation of Eluent Stock Solution Concentrates ". Make the required stock and working solutions for eluents.
3. See “Quality Assurance Report”. Run the Production Test Chromatogram as a system check.
4. See Section 5, "Example Applications" for example applications.
5. See “Column Care" for column cleanup and long-term storage recommendations.

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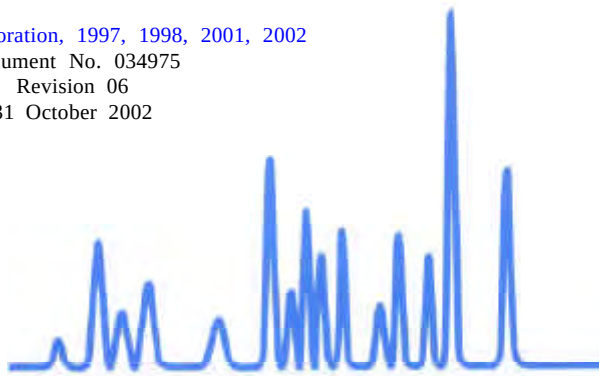


TABLE OF CONTENTS

SECTION 1 - INTRODUCTION TO IONPAC AS12A/AG12A CHROMATOGRAPHY

The IonPac® AS12A Analytical Column in combination with the AG12A Guard Column is designed for the analysis of fluoride and other inorganic anions including oxyhalides, such as chlorite and bromate. The selectivity of the IonPac AS12A Guard plus Analytical Column set has been designed to retain fluoride well out of the water dip (system dip) and to separate F^-, ClO_2^-, BrO_3, Cl^-, NO_2^-, Br^-, NO_3^-, HPO_4^{2-} and SO_4^{2-} isocratically in less than 12 minutes. The AS12A is compatible with pH 0-14 eluents and eluents containing organic solvents from 0 - 100% in concentration. The AS12A can be used with any suppressible ionic eluent that does not exceed the capacity of the suppressor. The IonPac AS12A has nominal efficiency for sulfate using standard operating conditions of at least 20,000 plates/meter.

CAUTION

Do not run deionized water through the column for longer than 15 minutes at 1 mL/min (4-mm systems) or 0.25 mL/min (2-mm systems).

Table 1
IonPac AS12A/AG12A Packing Specifications

Column	Particle Diameter µm	Substrate[a] X-linking %	Latex Diameter nm	Latex[b] X-Linking %	Column Capacity µeq/column	Functional Group	Hydophobicity
AS12A 4 x 200 mm	9.0	55	140	0.2	52	Alkyl quaternary ammonium	Medium
AG12A 4 x 50 mm	13.0	55	160	0.5	4	Alkanol quaternary ammonium	Medium - Low
AS12A 2 x 200 mm	9.0	55	140	0.2	13	Alkyl quaternary ammonium	Medium
AG12A 2 x 50 mm	13.0	55	160	0.5	1	Alkanol quaternary ammonium	Medium - Low

[a] macroporous (2,000 Å) divinylbenzene/ethylvinylbenzene polymer
[b] microporous polyvinylbenzylammonium polymer cross-linked with divinylbenzene

Table 2
AS12A/AG12A Operating Parameters

Column	Typical Back Pressure psi (MPa)	Standard Flow Rate mL/min	Maximum Flow Rate mL/min
AS12A 4-mm Analytical	≤ 1,650 (11.37)	1.5	2.0
AG12A 4-mm Guard	≤ 300 (2.06)	1.5	2.0
AS12A + AG12A 4-mm columns	**≤ 1,950 (13.43)**	**1.5**	**2.0**
AS12A 2-mm Analytical	≤ 1,800 (12.41)	0.38	0.5
AG12A 2-mm Guard	≤ 300 (2.06)	0.38	0.5
AS12A + AG12A 2-mm columns	**≤ 2,100 (15.47)**	**0.38**	**0.5**

Always remember that assistance is available for any problem that may be encountered during the shipment or operation of DIONEX instrumentation and columns through the DIONEX North America Technical Call Center at 1-800-DIONEX-0 (1-800-346-6390) or through any of the DIONEX Offices listed in, "DIONEX Worldwide Offices."

SECTION 2 - COMPARISON OF CHROMATOGRAPHY SYSTEMS

The proper configuration of 2-mm system injection volume, mass loading, system void volume and flow rate is based the ratio of the 2-mm to 4-mm column cross-sectional area which is a factor of 1/4.

CONFIGURATION	2-mm	4-mm
Eluent Flow Rate	0.38 mL/min	1.5 mL/min
SRS	ASRS-ULTRA (2-mm) (P/N 053947)	ASRS-ULTRA (4-mm) (P/N 053946)
MMS	AMMS III (2-mm) (P/N 056751)	AMMS III (4-mm) (P/N 056750)
AES	AAES (P/N 056116)	AAES (P/N 056116)
NOTE Do not run suppressors over 40°C. If application requires a higher temperature, place suppressor outside of chromatographic oven.		
Injection Loop	2 - 15 µL	10 - 50 µL
System Void Volume	Eliminate switching valves, couplers and the GM-3 Gradient Mixer. Use only the 2-mm GM-3 Mixer (P/N 043149).	Minimize dead volumes. Switching valves, couplers can be used. Use the GM-2 , GM-3 or recommended gradient mixers.
Pumps	Use the GS50/GP50/GP40/IP20/IP25 in Microbore Configuration with a Microbore GM-4 (2-mm) Gradient Mixer. No External Gradient Mixer is required for GS50/GP50/GP40 Pump when performing gradient analysis The GPM-2 can be used for 2-mm isocratic chromatography at flow rates of 0.5 mL/min or greater but cannot be used for 2-mm gradient chromatography	Use the GP40/GP50/IP20/IP25 in Standard-Bore Configuration. The GM-3 Gradient Mixer should be used for gradient analysis on systems other than the GP40/GP50/IP20/IP25 and the DX-300 HPLC Pump.

CONFIGURATION	2-mm	4-mm
Detectors	AD20/AD25 Cell (6-mm, 7.5 µL, P/N 046423) VDM-2 Cell (3-mm, 2.0 µL, P/N 043120) CD20, CD25, CD25A, ED40, ED50 or ED50A Conductivity Cell with DS3 P/N 044130 or Conductivity Cell with shield P/N 044132 CDM-2/CDM-3 Cell P/N 042770 Replace the TS-1 with the TS-2 (P/N 043117) on the CDM-2 or the CDM-3. The TS-2 has been optimized for 2-mm operation. Do not use the TS-2 or the TS-1 with the ED40/ED50/ED50A or the CD20/CD25/CD25A. DIONEX Back Pressure Regulator 75 psi rating (P/N 039760, 046480) or Tubing (see Table 3) Ensure 50-75 psi back pressure.	AD20/AD25 Cell (10-mm, 9 µL, P/N 049393) VDM-2 Cell (6-mm, 10 µL) P/N 043113 CD20, CD25, CD25A, ED40, ED50 or ED50A Conductivity Cell with DS3 P/N 044130 or with shield P/N 044132 CDM-2/CDM-3 Cell P/N 042770 Either the TS-1 with the TS-2 can be used with the CDM-2 or the CDM-3. Do not use the TS-2 or the TS-1 with the ED40/ED50/ED50A or the CD20/CD25/CD25A. DIONEX Back Pressure Regulator 75 psi rating (P/N 039760, 046480) or Tubing (see Table 3) Ensure 50-75 psi back pressure.

Table 3
Tubing Back Pressures

Tubing ID in	H_20 Back Pressure Psi/ft at 1 mL/min
0.005	111.4
0.007	29.0
0.010	7.0
0.012	3.4

SECTION 3 - INSTALLATION

3.1 System Requirements

3.1.1 System Requirements for 2-mm Operation

The IonPac AS12A 2-mm Guard and Analytical Columns are designed to be run on the following DIONEX Ion Chromatographs equipped with suppressed conductivity detection. Isocratic analyses at flow rates of 0.5 mL/min or greater can be performed on a GS50/GP50/GP40/IP25, Gradient Pump Module (GPM-2) or an Advanced Gradient Pump (AGP) with standard (1/8" pistons) pump heads. For isocratic analyses at flow rates below 0.5 mL/min and gradient analyses, a Microbore GS50/GP50/GP40 or Advanced Gradient Pump (1/16" pistons) must be employed.

3.1.2 System Requirements for 4-mm Operation

The IonPac AS12A 4-mm Guard and Analytical Columns are designed to be run on any DIONEX Ion Chromatograph equipped with suppressed conductivity detection. Gradient methods and methods requiring solvent containing eluents should be performed on a system having a Gradient Pump Module (GPM-2) or an Advanced Gradient Pump (AGP) or a GP40/GP50/GS50 with standard 1/8" pump heads. Isocratic analysis can also be performed on an IP20/IP25 with standard bore.

3.1.3 System Void Volume

When using 2-mm columns, it is particularly important to minimize system void volume. The system void volume should be scaled down to at least 1/4 of the system volume in a standard 4-mm system. For best performance, all of the tubing installed between the injection valve and detector should be 0.005" (P/N 044221) ID PEEK tubing. 0.010" ID PEEK tubing (P/N 042260) or 0.012" Tefzel tubing (see, “DIONEX Product Selection Guide”) may be used but peak efficiency will be compromised which may also result in decreased peak resolution. Minimize the lengths of all connecting tubing and remove all unnecessary switching valves and couplers. If you need assistance in properly configuring your system contact the DIONEX Office nearest you (see, “DIONEX Worldwide Offices”).

3.2 The Sample Concentrator

The Low Pressure Trace Anion Concentrator Column (TAC-LP1, P/N 046026), the Trace Anion Concentrator Column (TAC-2, P/N 043101), the IonPac AG12A Guard Column, or the Anion MicroConcentrator (AMC-1, P/N 051760) can be used for trace anion concentration work required in high purity water analysis. The function of the TAC-LP1, the TAC-2, the AMC-1, or the AG12A Guard Column in these applications is to strip ions from a measured volume of a relatively clean aqueous sample matrix. This process “concentrates” all anionic analyte species onto the TAC-LP1, TAC-2, AMC-1, or the AG12A leading to a lowering of detection limits by 2-5 orders of magnitude. The unique advantage to the analytical chemist of the TAC-LP1, the TAC-2, the AMC-1, or the AG12A in these applications is the capability of performing routine trace analyses of sample matrix ions at µg/L levels without extensive and laborious sample pretreatment.

For a detailed discussion of anion concentration techniques, refer to Section 3, “Operation,” of the Low Pressure Trace Anion Concentrator (TAC-LP1) Column Product Manual (Document No. 034972).

3.3 The Injection Loop

Table 4
Smallest Injectable Volumes (µL)

Valve Type	Using 0.012" ID Tefzel Tubing	Using 0.007" ID Tefzel Tubing	Using 0.010" ID PEEK Tubing	Using 0.005" ID PEEK Tubing
DIONEX BF2 Valve (8 µL Internal Volume) (10 cm Loop)	15.2	10.5	13.1	9.2
DIONEX MicroInject Valve (10.5 µL Internal Volume) (14 cm Loop)	20.5	14.0	17.6	12.2
Rheodyne Microinjection Valve Model 9126 (0.8 µL Internal Volume) (10 cm Loop)	8.0	3.3	5.9	2.0

3.3.1 The 2-mm System Injection Loop, 2 - 15 µL

For most applications on a 2-mm analytical system, a 2 - 15 µL injection loop is sufficient. DIONEX recommends that a 2.5 µL injection loop be used to avoid overloading the AS12A 2-mm Analytical Column. Generally, you should not inject more than 2.5 nanomoles (100 - 200 ppm) of any one analyte onto a 2-mm analytical column. Injecting larger volumes of samples can result in overloading the column which can affect the detection linearity. The AS12A 2-mm requires a microbore HPLC system configuration. Install an injection loop one-fourth or less (<15 µL) of the loop volume used with a 4-mm analytical system (Section 2, “Comparison of Chromatography Systems”).

3.3.2 The 4-mm System Injection Loop, 10 - 50 µL

For most applications on a 4-mm analytical system, a 10 - 50 µL injection loop will be sufficient. DIONEX recommends that a 10 µL injection loop be used to avoid overloading the AS12A 4-mm Analytical Column. Generally, do not inject more than 10 nanomoles (100 - 200 ppm) of any one analyte onto the 4-mm analytical column. Injecting larger volumes of samples can result in overloading the column which can affect the detection linearity. This phenomenon will be more prevalent at higher concentrations of the analytes of interest.

3.4 The IonPac AG12A Guard Column

An IonPac AG12A Guard Column is normally used with the IonPac AS12A Analytical Column. Retention times will increase by approximately 15% when a guard column is placed in-line prior to the analytical column. A guard is placed prior to the analytical column to prevent sample contaminants from eluting onto the analytical column. It is easier to clean or replace a guard column than it is an analytical column. Replacing the AG12A Guard Column at the first sign of peak efficiency loss or decreased retention time will prolong the life of the AS12A Analytical Column.

3.5 Installing the Anion Trap Column, ATC-3

When performing a gradient anion exchange application, a borate eluent system should be used instead of a carbonate system because of its low background conductivity. An IonPac Anion Trap Column (ATC-3 (2-mm), P/N 059661 or ATC-3 (4-mm), P/N 059660) should be installed between the Gradient Pump and the injection valve. Remove the high pressure Gradient Mixer if present. The ATC is filled with high capacity anion exchange resin which helps to minimize the baseline shift caused by increasing anionic contaminant levels in the eluent as the ionic concentration of the eluent is increased over the course of the gradient analysis.

To install the ATC-3 (2-mm) or ATC-3 (4-mm), complete the following steps:

A. Remove the Gradient Mixer, if installed between the gradient pump pressure transducer and the injection valve.

B. Connect the gradient pump directly to the ATC. Connect a waste line to the ATC outlet and direct the line to a waste container.

C. Flush the ATC with 200 mL of 70 mM $Na_2B_4O_7$ at a flow rate of 0.5 mL/min when using the ATC-3 (2-mm) or 2.0 mL/min when using the ATC-3 (4-mm).

D. Rinse the ATC with the strongest eluent that will be used during the gradient analysis.

E. After flushing the ATC with eluent, connect the ATC to the eluent line that is connected to the injection valve.

The background conductivity of your system should be less than 7 µS when $Na_2B_4O_7$ is being pumped through the chromatographic system with the ASRS in-line and properly functioning. The baseline shift should be no greater than 10 mS during a borate gradient eluent concentration ramp from 0 to 70 mM $Na_2B_4O_7$. If the baseline shifts are greater than 10 µS, the ATC should be cleaned using steps A - E above.

The ATC can be flushed, at the end of each operating day, to remove any impurities that may have accumulated on it. This will minimize periodic maintenance and lost data.

A. Flush the ATC with 30 mL of 70 mM $Na_2B_4O_7$.

B. Prior to next day use of the chromatographic system, flush the ATC with 30 mL of the strongest eluent used in the gradient program.

See the Product Manual for the IonPac ATC-3 (P/N 032697) for instructions on cleaning a contaminated Anion Trap Column.

At the end of each operating day, the ATC-3 should be flushed to remove any impurities that may have accumulated on it.

Under normal operating conditions, the ATC-3 column should be regenerated at the end of each operational day to remove any contaminants that may have collected on it, including carbonate. The daily regeneration of the ATC-3 column ensures that the IC system is systematically equilibrated for the most reproducible determinations of those anions being eluted by the weak eluents.

See the conditioning procedure above for regeneration of ATC-3 columns. For detailed information refer to the ATC-3 Product Manual (Document No. 032697).

3.6 Eluent Storage

IonPac AS12A columns are designed to be used with bicarbonate/carbonate eluent systems. Storage under a helium atmosphere ensures contamination free operation and proper pump performance (nitrogen can be used if eluents do not contain solvents).

3.7 Anion Self-Regenerating Suppressor Requirements

An Anion Self-Regenerating Suppressor should be used for applications that require suppressed conductivity detection. It is compatible with solvent containing eluents and aqueous ionic eluents of all concentrations with which the systems and columns are compatible. Aqueous ionic eluents can be used in all ASRS-ULTRA modes of operation.

NOTE
Solvent containing eluents should be used in the AutoSuppression External Water Mode.

If you are installing an IonPac AS12A 4-mm Analytical Column, use an ASRS-ULTRA (4-mm, P/N 053946).
If you are installing an IonPac AS12A 2-mm Analytical Column, use an ASRS-ULTRA (2-mm, P/N 053947).

For detailed information on the operation of the Anion Self-Regenerating Suppressor, see Document No. 031367, the "Product Manual for the Anion Self-Regenerating Suppressor-ULTRA, the ASRS-ULTRA."

3.8 Anion Atlas Electrolytic Suppressor Requirements

An Anion Atlas® Electrolytic Suppressor (AAES) may be used instead of an ASRS-ULTRA for applications that require suppressed conductivity detection. The AAES (P/N 056116) can be used for 2-mm and 4-mm IonPac AS12A applications using eluents up to 25 µeq/min.

For detailed information on the operation of the Anion Atlas Electrolytic Suppressor, see Document No. 031770, the "Product Manual for the Anion Atlas Electrolytic Suppressor."

3.9 Anion MicroMembrane Suppressor Requirements

An Anion MicroMembrane Suppressor (AMMS III) may be used instead of an ASRS-ULTRA (4-mm) for applications that require suppressed conductivity detection. Use an AMMS III (P/N 056750) with the IonPac AS12A 4-mm Analytical Column. It is compatible with all solvents and concentrations with which the systems and columns are compatible. For 2-mm operation, use the AMMS III (2-mm) (P/N 056751).

For detailed information on the operation of the Anion MicroMembrane Suppressor, see Document 031727, the "Product Manual for the Anion MicroMembrane Suppressor III, the AMMS III."

3.10 Using Displacement Chemical Regeneration (DCR) with the Chemical Suppression Mode

DIONEX recommends using the Displacement Chemical Regeneration (DCR) Mode for chemical suppression using sulfuric acid and the Anion MicroMembrane Suppressor (AMMS III). See the DCR kit manual, Document P/N 031664, for details.

SAFETY
Use proper safety precautions in handling acids and bases.

3.11 Using AutoRegen with the ASRS-ULTRA or the AMMS in the Chemical Suppression Mode

To save regenerant preparation time and reduce regenerant consumption and waste, DIONEX recommends using an AutoRegen® Accessory (P/N 039594). For more detailed information on the use of the AutoRegen Accessory see the AutoRegen Accessory manual (Document No. 032853). For more detailed information on the use of AutoRegen Regenerant Cartridges, see the "Product Manual for the AutoRegen Regenerant Cartridge Refills" (Document No. 032852).

3.12 Detector Requirements

See Section 2, "Comparison of 2-mm and 4-mm Ion Chromatography Systems," for 2-mm and 4-mm system detector, cell and thermal stabilizer requirements.

SECTION 4 - OPERATION

4.1 General Operating Conditions

Sample Volume:	2-mm: 2.5 µL Loop + 0.8 µL Injection valve dead volume 4-mm: 10 µL Loop + 0.8 µL Injection valve dead volume
Column:	2-mm: AS12A 2-mm Analytical Column + AG12A 2-mm Guard Column 4-mm: AS12A 4-mm Analytical Column + AG12A 4-mm Guard Column
Eluent:	2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Eluent Flow Rate:	2-mm: 0.38 mL/min 4-mm: 1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (2-mm or 4-mm) AutoSuppression Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (2-mm or 4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS
Storage Solution:	0.1 M NaOH

4.2 IonPac AS12A Operation Precautions

CAUTION
Filter and Degas Eluents
Filter Samples
Eluent pH between 0 and 14
Sample pH between 0 and 14
0.5 mL/min Maximum Flow Rate for 2-mm Columns
2.0 mL/min Maximum Flow Rate for 4-mm Columns
Maximum Operating Pressure = 3,500 psi (24.13 MPa)

4.3 Chemical Purity Requirements

Obtaining reliable, consistent and accurate results requires eluents that are free of ionic impurities. Chemicals, solvents and deionized water used to prepare eluents must be of the highest purity available. Low trace impurities and low particle levels in eluents also help to protect your ion exchange columns and system components. DIONEX cannot guarantee proper column performance when the quality of the chemicals, solvents and water used to prepare eluents has been compromised.

4.3.1 Inorganic Chemicals

Reagent Grade inorganic chemicals should always be used to prepare ionic eluents. Whenever possible, inorganic chemicals that meet or surpass the latest American Chemical Society standard for purity should be used. These inorganic chemicals will detail the purity by having an actual lot analysis on each label.

4.3.2 Deionized Water

The deionized water used to prepare eluents should be Type I Reagent Grade Water with a specific resistance of 18.2 megohm-cm. The deionized water should be free of ionized impurities, organics, microorganisms and particulate matter larger than 0.2 µm Bottled HPLC-Grade Water (with the exception of Burdick & Jackson) should not be used since most bottled water contains an unacceptable level of ionic impurities.

4.3.3 Solvents

Solvents can be added to the ionic eluents used with IonPac AS12A columns to modify the ion exchange process or improve sample solubility. The solvents used must be free of ionic impurities. However, since most manufacturers of solvents do not test for ionic impurities, it is important that the highest grade of solvents available be used. Currently, several manufacturers are making ultrahigh purity solvents that are compatible for HPLC and spectrophotometric applications. These ultrahigh purity solvents will usually ensure that your chromatography is not affected by ionic impurities in the solvent. Currently at DIONEX, we have obtained consistent results using High Purity Solvents manufactured by Burdick and Jackson and Optima® Solvents by Fisher Scientific.

When using a solvent in an ionic eluent, column generated back pressures will depend on the solvent used, concentration of the solvent, the ionic strength of the eluent and the flow rate used. The column back pressure will vary as the composition of water-methanol and water-acetonitrile mixture varies. The practical back pressure limit for the IonPac AS12A columns is 3,500 psi.

The AS12A can withstand common HPLC solvents in a concentration range of 0 - 100%. Solvents and water should be premixed in concentrations which allow proper mixing by the gradient pump and to minimize outgassing. Ensure that all of the inorganic chemicals are soluble in the highest solvent concentration to be used during the analysis.

Table 5
HPLC Solvents for Use with IonPac AS12A Columns

Solvent	Maximum Operating Concentration
Acetonitrile	100%
Methanol	100%
2-Propanol	100%
Tetrahydrofuran	20%

CAUTION

The Anion Self-Regenerating Anion Suppressor (ASRS-ULTRA) must be operated in the AutoSuppression External Water Mode when using eluents containing solvents.

4.4 Making Eluents that Contain Solvents

When mixing solvents with water remember to mix solvent with water on a volume to volume basis. If a procedure requires an eluent of 90% acetonitrile, prepare the eluent by adding 900 mL of acetonitrile to an eluent reservoir. Then add 100 mL of deionized water or eluent concentrate to the acetonitrile in the reservoir. Using this procedure to mix solvents with water will ensure that a consistent true volume/volume eluent is obtained. Premixing water with solvent will minimize the possibility of outgassing.

NOTE

When purging or degassing eluents containing solvents, do not purge or degas the eluent excessively since it is possible that a volatile solvent can be “boiled” off from the solution.

Always degas and store all eluents in glass or plastic eluent bottles pressurized with helium. Only helium can be used to purge and degas ionic eluents containing solvents, since nitrogen is soluble in solvent containing eluents.

Acetonitrile (ACN) hydrolyzes to ammonia and acetate when left exposed to basic solutions. To prevent eluent contamination from acetonitrile hydrolysis, always add acetonitrile to basic aqueous eluents by proportioning the acetonitrile into the basic eluent with the gradient pump. Keep the acetonitrile in a separate eluent bottle containing only acetonitrile and water.

CAUTION
Never add the acetonitrile directly to the basic carbonate or hydroxide eluent bottle.

4.5 Regenerant Preparation for the AMMS III

The Anion MicroMembrane Suppressor III (AMMS III) requires the use of a regenerant solution. If you are using the AMMS III instead of the Anion Self-Regenerating Suppressor-ULTRA (ASRS-ULTRA) see Document No. 031727, the "Product Manual for the Anion MicroMembrane Suppressor III, the AMMS III."

4.6 Sample Concentration

The Low Pressure Trace Anion Concentrator (TAC-LP1), the Trace Anion Concentrator (TAC-2), the Anion MicroConcentrator (AMC-1), or the IonPac AG12A Guard Column can be used for trace anion concentration work required in high purity water analysis. The function of a concentrator column in these applications is to strip ions from a measured volume of a relatively clean aqueous sample matrix. This process "concentrates" the desired analyte species onto the concentrator column, lowering detection limits by 2-5 orders of magnitude. The concentrator column is used in lieu of the sample loop. Pump the sample onto the concentrator column in the **OPPOSITE** direction of the eluent flow. When using concentration techniques, do not overload the concentrator column by concentrating an excessive amount of sample. Concentrating an excessive amount of sample can result in inaccurate results being obtained. It is possible during the concentration step for the polyvalent anions such as phosphate and sulfate to elute the weakly retained anions such as fluoride and acetate off the concentrator column. For more detailed information on sample concentration techniques for high sensitivity work refer to Section 3, "Operation," of the Low Pressure Trace Anion Concentrator (TAC-LP1) Column Product Manual (Document No. 034972). These techniques can also be applied to either the TAC-2, AMC-1, or the AG12A.

SECTION 5 - EXAMPLE APPLICATIONS

The chromatograms in this section were obtained using columns that reproduced the Production test Chromatogram (see Section 5.2, "Production Test Chromatogram") on optimized Ion Chromatographs (see Section 3, "Installation"). Different systems will differ slightly in performance due to slight differences in column sets, system void volumes, liquid sweep-out times of different components and laboratory temperatures.

Before attempting any of the following example applications, take the time to ensure that your system is properly configured. Ensure that all of the eluents have been made from high purity reagents and deionized water. All water used in the preparation of eluents should be degassed, deionized water. For chemical purity requirements, see Section 4.3, "Chemical Purity Requirements." After running synthetic standards to calibrate your system, you may find that real sample matrices foul your columns. For this reason it is always advisable to use a guard column to protect the analytical column. If column performance deteriorates and it is determined that the guard or the analytical column has been fouled, refer to the column cleanup protocols in, "Column Care." If your sample matrices are relatively low in ionic concentration, you may be able to increase the sensitivity of your system by using sample concentration techniques (see Section 4.6, "Sample Concentration").

5.1 Preparation of Eluent Stock Solution Concentrates

A. 0.5 M Sodium Carbonate (Na_2CO_3) Concentrate

Order DIONEX P/N 037162
or
Thoroughly dissolve 26.49 g of Na_2CO_3 in 400 mL of deionized water with a specific resistance of 18.2 megohm-cm. Dilute to a final volume of 500 mL.

B. 0.5 M Sodium Bicarbonate ($NaHCO_3$) Concentrate

Order DIONEX P/N 037163
or
Thoroughly dissolve 21.00 g of $NaHCO_3$ in 400 mL of deionized water with a specific resistance of 18.2 megohm-cm. Dilute to a final volume of 500 mL.

5.2 Eluent Preparation

Eluent: 2.7 mM Sodium Carbonate/0.3 mM Sodium Bicarbonate

Prepare the eluent by pipetting 5.4 mL of 0.5 M Na_2CO_3 plus 0.6 mL of 0.5 M $NaHCO_3$ into a 1 L volumetric flask. Use degassed, deionized water with a specific resistance of 18.2 megohm-cm to dilute the concentrate to a final volume of 1,000 mL.

5.3 Production Test Chromatogram

Isocratic elution of anions on the IonPac AS12A Analytical Column has been optimized utilizing a carbonate/bicarbonate eluent. By using this eluent, mono- and divalent anions can be isocratically separated and quantitated in a single injection. The IonPac AS12A Analytical Column should always be used with the IonPac AG12A Guard Column. To guarantee that all IonPac AS12A Analytical Columns meet high quality and reproducible performance specification standards, all columns undergo the following production control test.

Sample Volume:	2-mm: 2.5 µL Loop + 0.8 µL Injection valve dead volume 4-mm: 10 µL Loop + 0.8 µL Injection valve dead volume
Column:	See Chromatogram
Eluent:	2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Eluent Flow Rate:	0.38 mL/min (2-mm), 1.5 mL/min (4-mm)
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS -ULTRA (2-mm or 4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (2-mm or 4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS
Storage Solution:	0.1 M NaOH

	Analyte	mg/L
1.	Fluoride	3.0
2.	Chlorite	20.0
3.	Bromate	20.0
4.	Chloride	6.0
5.	Nitrite	10.0
6.	Bromide	20.0
7.	Chlorate	20.0
8.	Nitrate	20.0
9.	Phosphate	30.0
10.	Sulfate	20.0

where 1 mg/L = 1 ppm

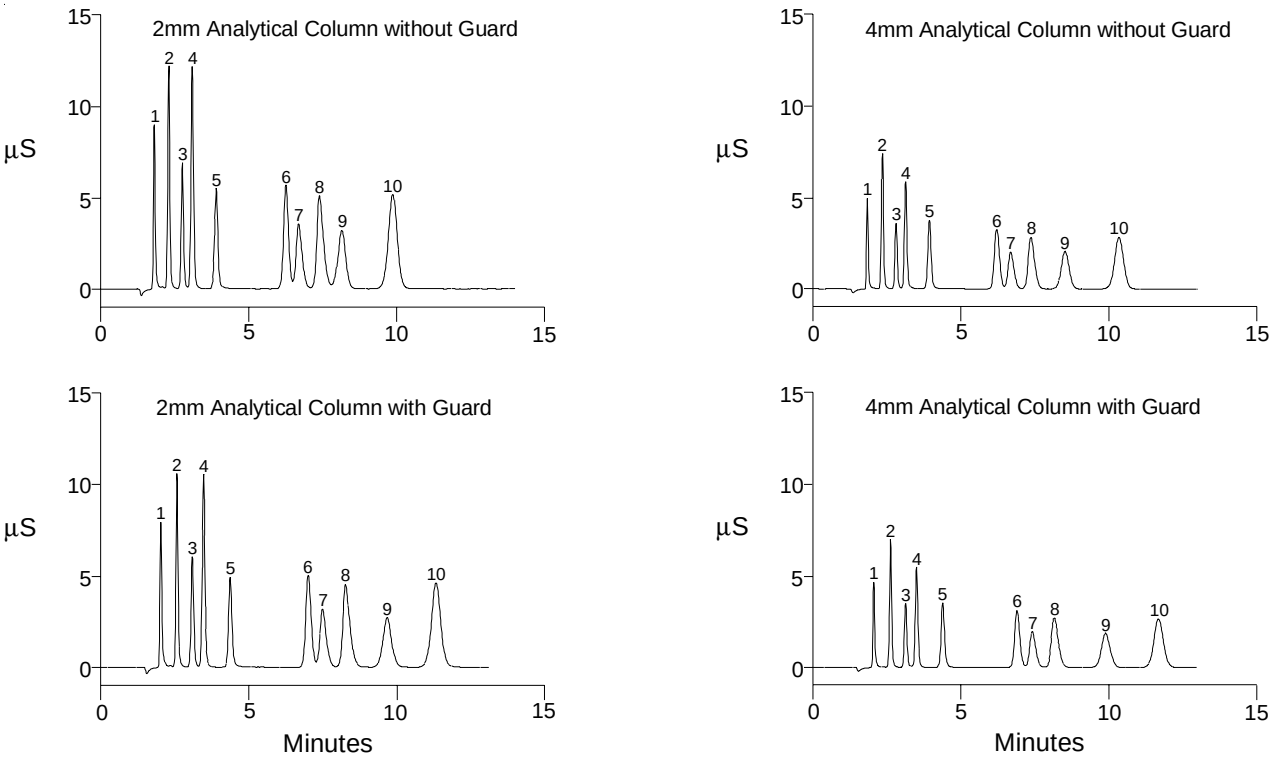


Figure 1
IonPac AS12A Production Test Chromatograms

5.4 Fast Run Analysis without Changes in Selectivity

The following chromatograms demonstrate the development of a fast run through eluent and flow rate changes that do not change the original selectivity.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column, 4-mm
Eluent:	See Chromatogram
Eluent Flow Rate:	See Chromatogram
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (2-mm or 4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (2-mm or 4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

	Analyte	mg/L
1.	Fluoride	3.0
2.	Chloride	5.0
3.	Nitrite	10.0
4.	Bromide	20.0
5.	Nitrate	20.0
6.	Phosphate	30.0
7.	Sulfate	20.0

where 1 mg/L = 1 ppm

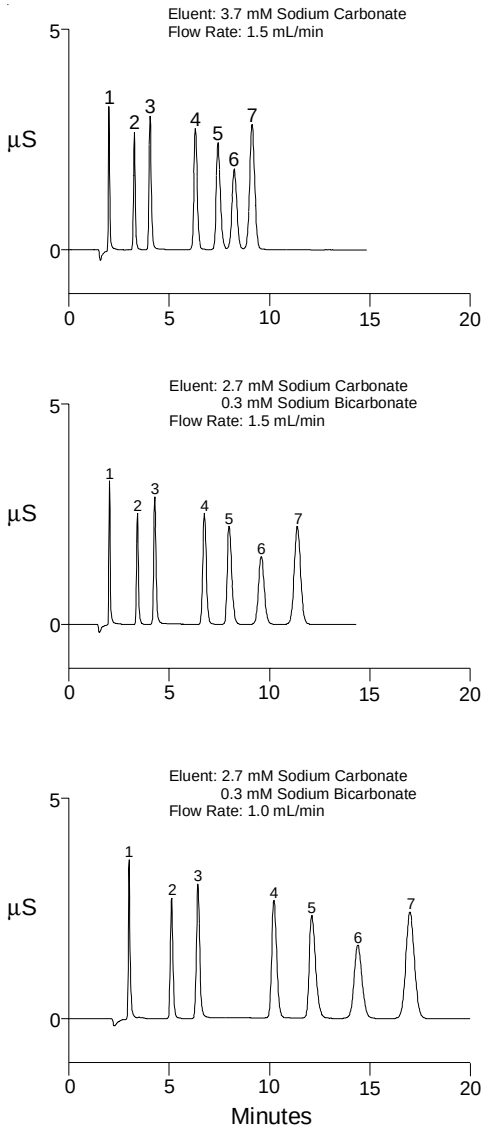


Figure 2
Fast Run Analysis Without Changes in Selectivity

5.5 Fast Run Analysis With Selectivity Changes

The following chromatogram demonstrates the elution of the 10 anions on the test chromatogram under fast run conditions. The run was obtained by increasing the flow rate and increasing the concentration of carbonate to increase both the ionic strength and the pH of the eluent. Note that with the change in the eluent, phosphate and sulfate move forward in the chromatogram to the area between nitrite and bromide.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column
Eluent:	10.5 mM Na_2CO_3/0.5 mM $NaHCO_3$
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

	Analyte	mg/L
1.	Fluoride	3.0
2.	Chlorite	20.0
3.	Bromate	20.0
4.	Chloride	5.0
5.	Nitrite	10.0
6.	Phosphate	30.0
7.	Sulfate	20.0
8.	Bromide	20.0
9.	Chlorate	20.0
10.	Nitrate	20.0

where 1 mg/L = 1 ppm

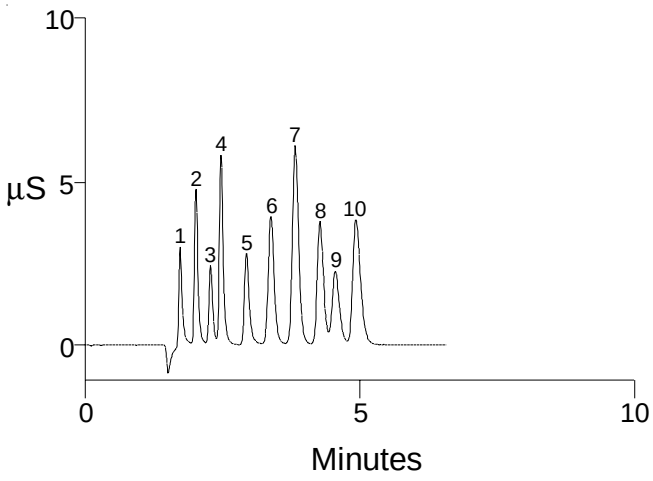


Figure 3
Fast Run Analysis With Selectivity Changes

5.6 Analysis of Acetate and Formate

The following chromatogram demonstrates the elution of acetate and formate along with the 10 anions on the test chromatogram using standard eluent conditions.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column, 4-mm
Eluent:	2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Eluent Flow Rate:	1.0 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

	Analyte	mg/L
1.	Fluoride	0.5
2.	Acetate	10.0
3.	Formate	5.0
4.	Chlorite	5.0
5.	Bromate	5.0
6.	Chloride	1.0
7.	Nitrite	2.0
8.	Bromide	5.0
9.	Chlorate	5.0
10.	Nitrate	5.0
11.	Phosphate	10.0
12.	Sulfate	5.0

where 1 mg/L = 1 ppm

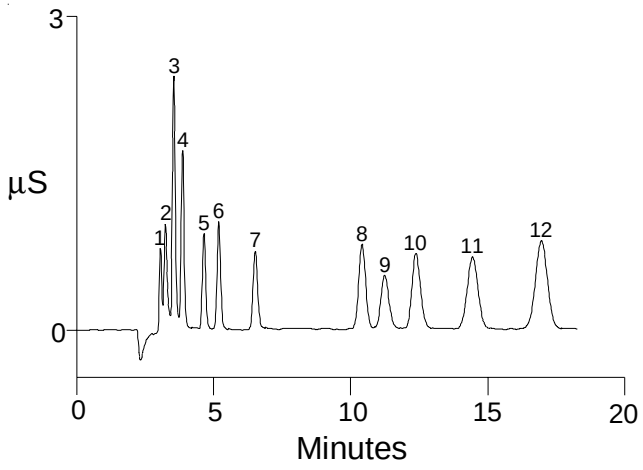


Figure 4
Analysis of Acetate and Formate

5.7 Analysis of Sulfite, Selenite, Selenate and Arsenate

The elution of sulfite, selenite, selenate and arsenate require eluent modification to avoid co-elution with other common anions. The following chromatogram demonstrates the eluent adjustment required to obtain resolution of these analytes.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column, 4-mm
Eluent:	See Chromatogram
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

	Analyte	mg/L
1.	Fluoride	3.0
2.	Chloride	5.0
3.	Nitrite	10.0
4.	Bromide	20.0
5.	Nitrate	20.0
6.	Selenite	20.0
7.	Phosphate	30.0
8.	Sulfite	20.0
9.	Sulfate	20.0
10.	Arsenate	40.0
11.	Selenate	20.0

where 1 mg/L = 1 ppm

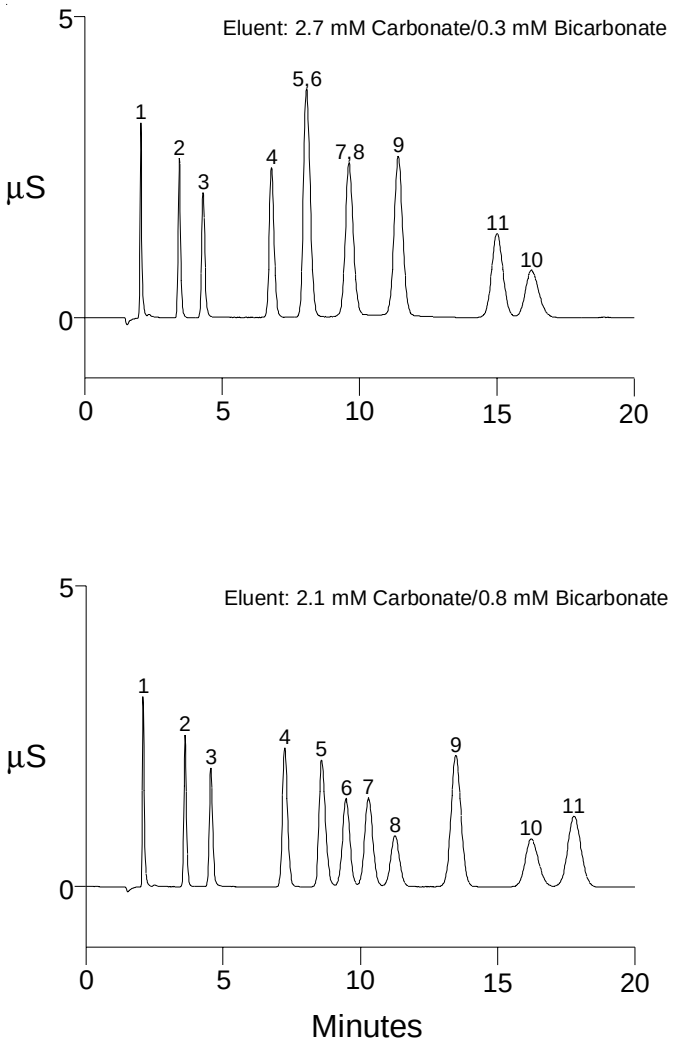


Figure 5
Analysis of Sulfite, Selenite, Selenate and Arsenate

5.8 Analysis of a Simulated Drinking Water Sample with a Carbonate Eluent

The following simulated drinking water sample demonstrates the types and concentrations of analytes found in typical drinking water samples.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column, 4-mm
Eluent:	2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

	Analyte	mg/L
1.	Fluoride	0.1
2.	Bicarbonate	20.0
3.	Chlorite	0.1
4.	Bromate	0.1
5.	Chloride	20.0
6.	Nitrite	0.1
7.	Bromide	0.1
8.	Chlorate	0.1
9.	Nitrate	5.0
10.	Phosphate	0.2
11.	Sulfate	20.0

where 1 mg/L = 1 ppm

	Analyte	mg/L
1.	Fluoride	3.0
2.	Bicarbonate	0.0
3.	Chlorite	20.0
4.	Bromate	20.0
5.	Chloride	5.0
6.	Nitrite	10.0
7.	Bromide	20.0
8.	Chlorate	20.0
9.	Nitrate	20.0
10.	Phosphate	30.0
11.	Sulfate	20.0

where 1 mg/L = 1 ppm

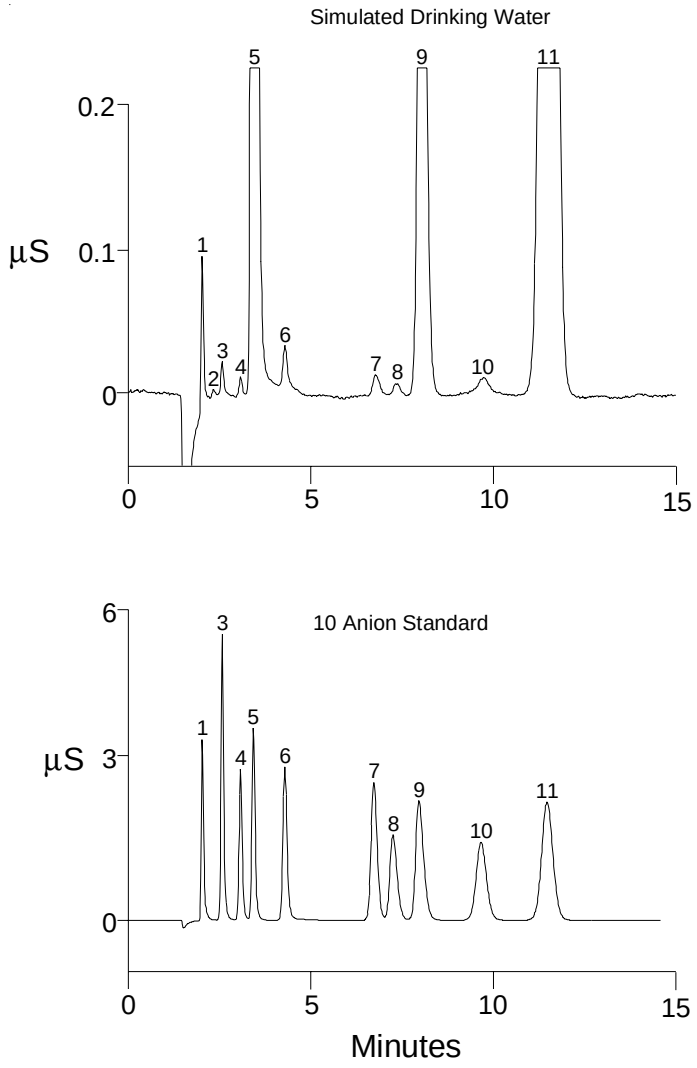


Figure 6
Analysis of a Simulated Drinking Water Sample with a Carbonate Eluent

5.9 Analysis of Bottled Drinking Water

The following analyses of bottled drinking water demonstrate typical concentrations of inorganic anions.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column, 4-mm
Eluent:	2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

Analyte
1. Fluoride
2. Carbonate
3. Chloride
4. Nitrate
5. Sulfate

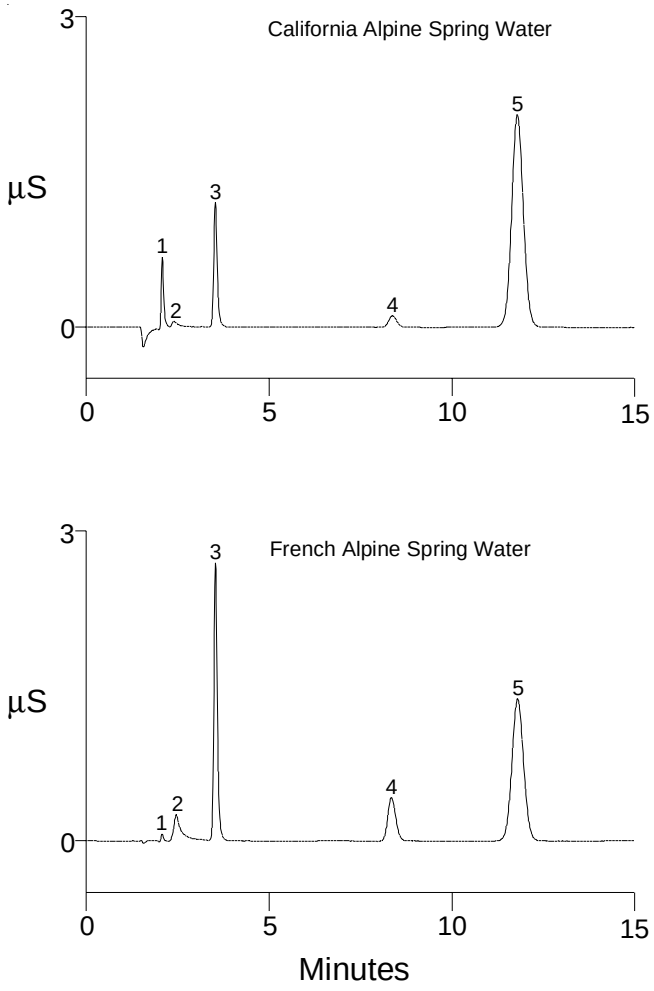


Figure 7
Analysis of Bottled Drinking Water

5.10 Analysis of City Tap Water

The following analyses of city tap water demonstrate typical types and concentrations of inorganic anions.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column, 4-mm
Eluent:	2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

Analyte
1. Fluoride
2. Carbonate
3. Chloride
4. Nitrate
5. Phosphate
6. Sulfate

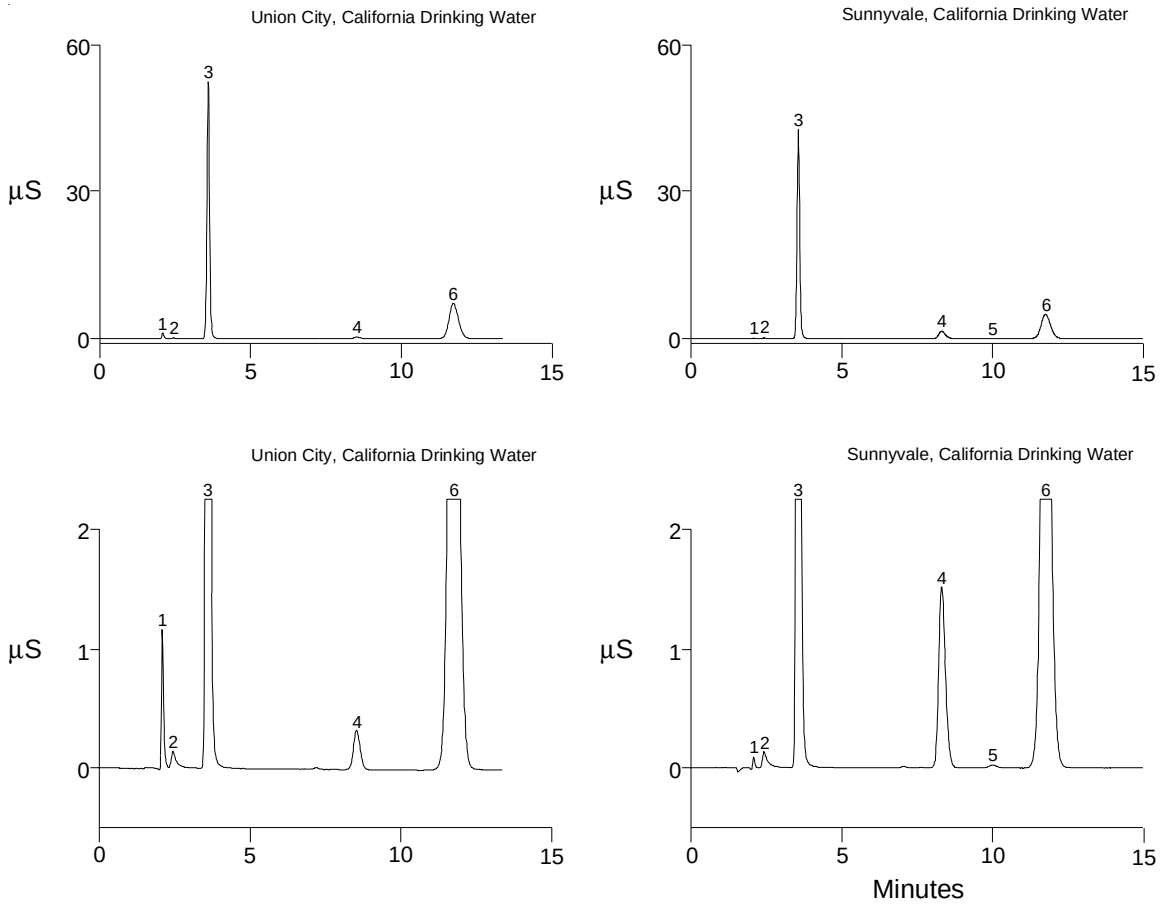


Figure 8
Analysis of City Tap Water

5.11 Analysis of a Simulated Drinking Water Sample with a Borate Eluent

The following simulated drinking water sample demonstrates the types and concentrations of analytes found in typical drinking water samples. Note the change in selectivity compared to the analysis done with the carbonate eluent (see Section 5.8, "Analysis of a Simulated Drinking Water Sample with a Carbonate Eluent"). The ASRS-ULTRA should be used in the Chemical Supperssion mode when using borate eluents.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column, 4-mm
Eluent:	18 mM NaOH/20 mM $Na_2B_4O_7$
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) Chemical Suppression Mode
SRS Regenerant:	35 mN H_2SO_4
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Regenerant Flow Rate:	8 mL/min.
Expected Background Conductivity:	3-5 µS

	Analyte	mg/L
1.	Fluoride	0.2
2.	Chlorite	0.2
3.	Bromate	0.2
4.	Chloride	40.0
5.	Bicarbonate	150.0
6.	Nitrite	0.2
7.	Phosphate	0.4
8.	Bromide	0.2
9.	Chlorate	0.2
10.	Nitrate	10.0
11.	Sulfate	40.0

where 1 mg/L = 1 ppm

	Analyte	mg/L
1.	Fluoride	3.0
2.	Chlorite	20.0
3.	Bromate	20.0
4.	Chloride	5.0
5.	Bicarbonate	150.0
6.	Nitrite	10.0
7.	Phosphate	30.0
8.	Bromide	20.0
9.	Chlorate	20.0
10.	Nitrate	20.0
11.	Sulfate	20.0

where 1 mg/L = 1 ppm

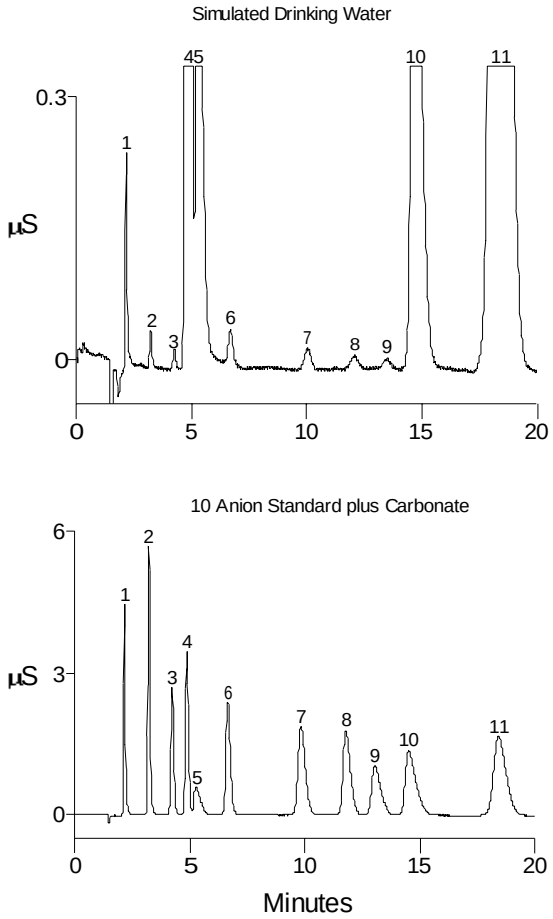


Figure 9
Analysis of a Simulated Drinking Water Sample with a Borate Eluent

5.12 Borate Gradient Analysis

The following analyses demonstrate the advantages of gradient analysis. Borate gradient elution enhances the resolution of the weakly retained analytes, allows better positioning of carbonate and accelerates the elution of the strongly retained analytes. The ASRS-ULTRA should be used in the Chemical Supperssion mode when using borate eluents.

Sample Loop Volume:	25 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column, 4-mm
Trap Column:	ATC-3, 4-mm
Isocratic Eluent:	18 mM NaOH/20 mM $Na_2B_4O_7$
Gradient Eluents:	E1: 37.5 mM NaOH/50 mM $Na_2B_4O_7$
	E2: Deionized water
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm)
	Chemical Suppression Mode
or SRS Regenerant:	35 mN H_2SO_4
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Regenerant Flow Rate:	8 mL/min.
Expected Background Conductivity:	3-5 µS

Gradient Conditions

Time	%E1	%E2	Comments
Init	22	78	Initial Eluent
0.00	22	78	Load Position
0.10	22	78	Inject
0.30	22	78	Load Position
16.00	73	27	End of Gradient
16.10	22	78	Reset Gradient

	Analyte	mg/L
1.	Fluoride	3.0
2.	Acetate	20.0
3.	Formate	20.0
4.	Chlorite	20.0
5.	Bromate	20.0
6.	Chloride	5.0
7.	Bicarbonate	150.0
8.	Nitrite	10.0
9.	Phosphate	30.0
10.	Bromide	20.0
11.	Chlorate	20.0
12.	Nitrate	20.0
13.	Sulfate	20.0

where 1 mg/L = 1 ppm

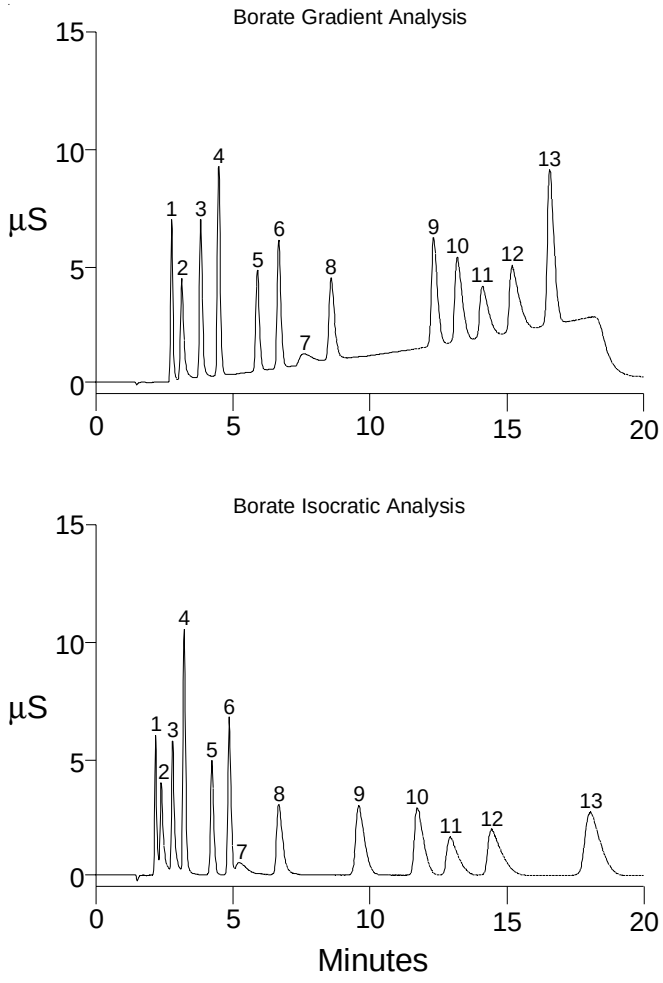


Figure 10
Borate Gradient Analysis

5.13 Anions in Tooth Paste

The following analyses demonstrate the quantification of monofluorophosphate in tooth paste. In each case, 1 g of sample was diluted to 100 g total weight with deionized water and then filtered first through a 0.45-µm syringe filter and then through a 0.20-µm syringe filter before injection.

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column+ IonPac AG12A Guard Column
Eluent:	2.5 mM Na_2CO_3/1.0 mM NaOH
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

	Analyte	mg/L
1.	Fluoride	3.0
2.	Chlorite	20.0
3.	Bromate	20.0
4.	Chloride	6.0
5.	Nitrite	10.0
6.	Bromide	20.0
7.	Chlorate	20.0
8.	Nitrate	20.0
9.	Monofluorophosphate	30.0
10.	Sulfate	20.0
11.	Phosphate	30.0

where:
1 mg/L = 1 ppm
u = Unknown

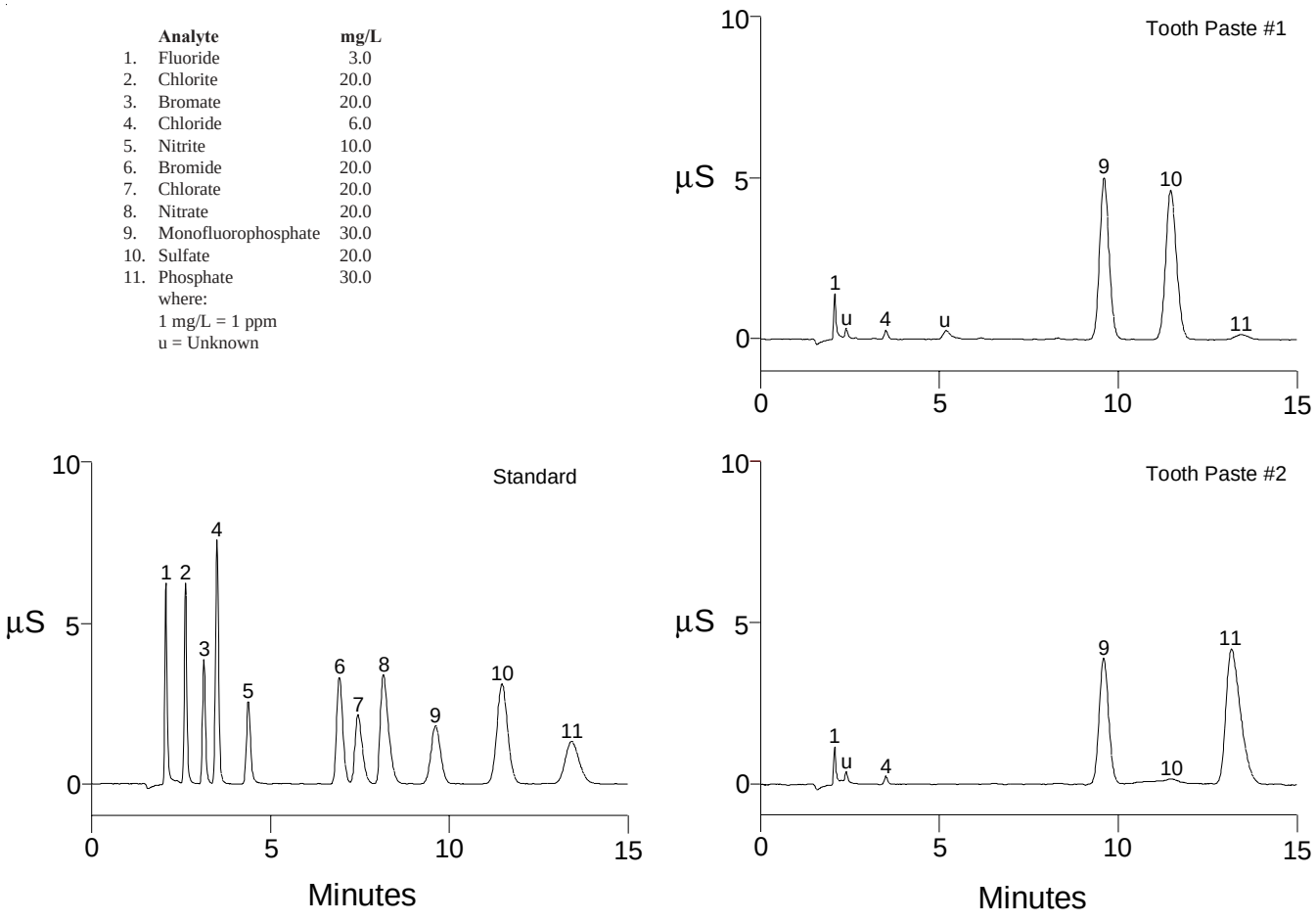


Figure 11
Anions in Tooth Paste

5.14 Kraft Black Liquor Fouling and Cleanup

The following analyses demonstrate the ability to clean the column after significant fouling with samples containing large quantities of hydrophobic organic acids..

Sample Loop Volume:	10 µL
Column:	IonPac AS12A Analytical Column, 4-mm
Eluent:	2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Eluent Flow Rate:	1.5 mL/min
SRS Suppressor:	Anion Self-Regenerating Suppressor, ASRS-ULTRA (4-mm) AutoSuppression® Recycle Mode
or AES Suppressor:	Anion Atlas Electrolytic Suppressor, AAES
or MMS Suppressor:	Anion MicroMembrane Suppressor, AMMS III (4-mm)
MMS Regenerant:	50 mN H_2SO_4
Expected Background Conductivity:	14-16 µS

	Analyte	mg/L
1.	Fluoride	3.0
2.	Chlorite	20.0
3.	Bromate	20.0
4.	Chloride	6.0
5.	Nitrite	10.0
6.	Bromide	20.0
7.	Chlorate	20.0
8.	Nitrate	20.0
9.	Phosphate	30.0
10.	Sulfate	20.0

where 1 mg/L = 1 ppm

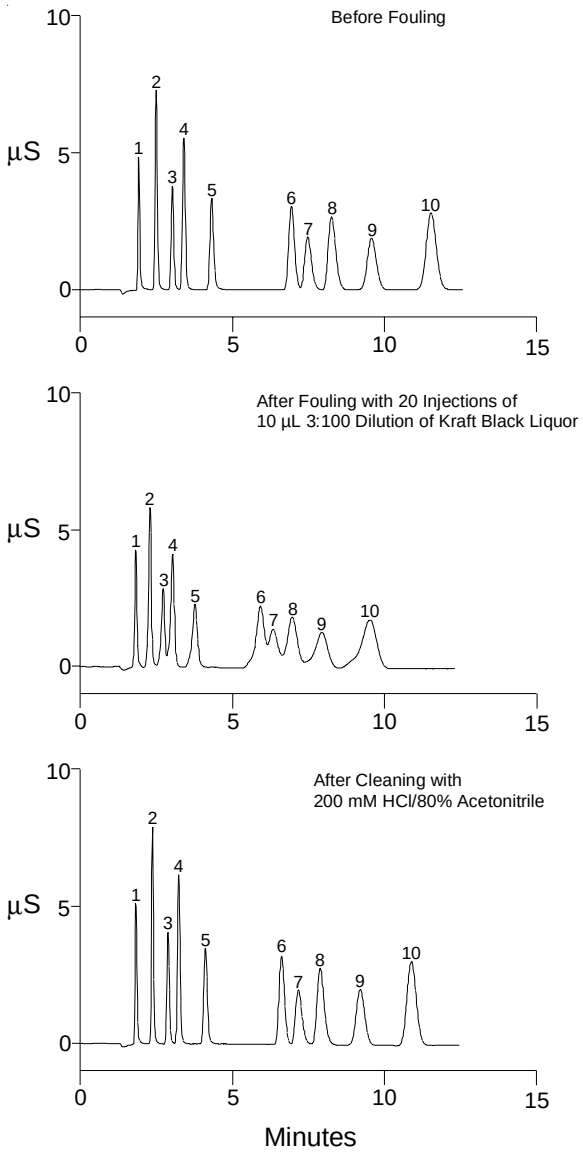


Figure 12
Kraft Black Liquor Fouling and Cleanup

SECTION 6 - TROUBLESHOOTING GUIDE

The purpose of the Troubleshooting Guide is to help you solve operating problems that may arise while using IonPac AS12A columns. For more information on problems that originate with the Ion Chromatograph (IC) or the suppressor, refer to the Troubleshooting Guide in the appropriate operator's manual. If you cannot solve the problem on your own, call the DIONEX Office nearest you (see, "DIONEX Worldwide Offices").

Table 6
AS12A/AG12A Troubleshooting Summary

Observation	Cause	Action	Reference Section
High Back Pressure	Unknown	Isolate Blocked Component	6.1.1
	Plugged Column Bed Supports	Replace Bed Supports	6.1.2
	Other System Components	Unplug, Replace	Component Manual
High Background Conductivity	Bad Eluents	Remake Eluents	6.2, 6.2.1
	Contaminated Columns	Clean Column	6.2.2, 6.2.3, Column Care
	Contaminated ASRS, AMMS, AAES	Clean Suppressor	6.2.5, 6.2.6, 6.2.7 Component Manual
	Contaminated Hardware	Clean Component	Component Manual
Poor Resolution	Poor Efficiency Due to Large System Void Volumes	Replumb System	6.3.1 B, Component Manual
	Column Headspace	Replace Column	6.3.1 A
Short Retention Times	Flow Rate Too fast	Recalibrate Pump	6.3.2 A
	Bad Eluents	Remake Eluents	6.3.2 B
	Column Contamination	Clean Column	6.3.2 C, 6.3.2 D, Column Care
Poor Front End Resolution	Bad Eluents	Remake Eluents	6.3.3 A
	Column Overloading	Reduce Sample Size	6.3.3 B, 3.3.1, 3.3.2
	Sluggish Injection Valve	Service Valve	6.3.3 C, Component Manual
	Large System Void Volumes	Replumb System	6.3.3 D, Component Manual
Spurious Peaks	Sample Contaminated	Pretreat Samples	6.3.4 A, 6.3.4 B, Column Care
	Sluggish Injection Valve	Service Valve	6.3.3 C, Component Manual

6.1 High Back Pressure

6.1.1 Finding the Source of High System Pressure

Total system pressure for the IonPac AG12A (4-mm) Guard Column plus the AS12A (4-mm) Analytical Column when using the test chromatogram conditions should be equal or less than 2,000 psi. If the system pressure is higher than 2,000 psi, it is advisable to determine the cause of the high system pressure. The system should be operated with a High-Pressure In-Line Filter (P/N 044105) which is positioned between the Gradient Pump pressure transducer and the injection valve. Make sure you have one in place and that it is not contaminated.

A. Make sure that the pump is set to the correct eluent flow rate. Higher than recommended eluent flow rates will cause higher pressure. Measure the pump flow rate if necessary with a stop watch and graduated cylinder.

B. Determine which part of the system is causing the high pressure. High pressure could be due to a plugged tubing or tubing with collapsed walls, an injection valve with a clogged port, a column with particulates clogging the bed support, a clogged High-Pressure In-Line Filter, the suppressor or the detector cell.

To determine which part of the chromatographic system is causing the problem, disconnect the pump eluent line from the injection valve and turn the pump on. Watch the pressure; it should not exceed 50 psi. Continue adding system components (injection valve, column(s), suppressor and detector) one by one, while monitoring the system pressure. The pressure should increase up to a maximum when the Guard and Analytical columns are connected (see Table 7, "Typical AS12A/AG12A Operating Back Pressures").

The suppressor may add up to 100 psi (0.69 MPa). No other components should add more than 100 psi (0.69 MPa) of pressure. Refer to the appropriate manual for cleanup or replacement of the problem component.

Table 7
Typical AS12A/AG12A Operating Back Pressures

Column	Typical Back Pressure psi (MPa)	Flow Rate mL/min
AS12A 4-mm Analytical	≤ 1,650 (11.37)	1.5
AG12A 4-mm Guard	≤ 300 (2.06)	1.5
AS12A + AG12A 4-mm columns	**≤ 1,950 (13.43)**	**1.5**
AS12A 2-mm Analytical	≤ 1,800 (12.41)	0.38
AG12A 2-mm Guard	≤ 300(2.06)	0.38
AS12A + AG12A 2-mm columns	**≤ 2,100 (15.47)**	**0.38**

6.1.2 Replacing Column Bed Support Assemblies

If the column inlet bed support is determined to be the cause of the high back pressure, it should be replaced. To change the inlet bed support assembly, refer to the following instructions, using one of the two spare inlet bed support assemblies included in the Ship Kit.

A. Disconnect the column from the system.

B. Carefully unscrew the inlet (top) column fitting. Use two open-end wrenches.

C. Remove the bed support. Turn the end fitting over and tap it against a benchtop or other hard, flat surface to remove the bed support and seal assembly. If the bed support must be pried out of the end fitting, use a sharp pointed object such as a pair of tweezers, but be careful that you **DO NOT SCRATCH THE WALLS OF THE END FITTING.** Discard the old bed support assembly.

D. Place a new bed support assembly into the end fitting. Make sure that the end of the column tube is clean and free of any particulate matter so that it will properly seal against the bed support assembly. Use the end of the column to carefully start the bed support assembly into the end fitting.

Product Descriptions	IonPac AS12A 4-mm Columns (P/N)	IonPac AS12A 2-mm Columns (P/N)
Analytical Column	046034	046055
Guard Column	079801	046056
Bed Support Assembly	042955	044689
End Fitting	052809	043278

CAUTION

If the column tube end is not clean when inserted into the end fitting, particulate matter may obstruct a proper seal between the end of the column tube and the bed support assembly. If this is the case, additional tightening may not seal the column but instead damage the column tube or the end fitting.

E. Screw the end fitting back onto the column. Tighten it fingertight, then an additional 1/4 turn (25 in x lb). Tighten further only if leaks are observed.

F. Reconnect the column to the system and resume operation.

NOTE

Replace the outlet bed support ONLY if high pressure persists after replacement of the inlet fitting.

6.2 High Background or Noise

In a properly working system, the background conductivity level for the standard eluent system is shown below:

ELUENT	EXPECTED BACKGROUND CONDUCTIVITY
2.7mM Na_2CO_3/0.3 mM $NaHCO_3$	14 - 16 µS
18 mM NaOH/20 mM $Na_2B_4O_7$	3 - 5 µS

6.2.1 Preparation of Eluents

A. Make sure that the eluents and the regenerant are made correctly.

B. Make sure that the eluents are made from chemicals with the recommended purity.

C. Make sure that the deionized water used to prepare the reagents has a specific resistance of 18.2 megohm-cm.

6.2.2 A Contaminated Anion Trap Column, ATC-3

When doing gradient analysis, has the Anion Trap Column, the ATC-3 (2-mm) or the ATC-3 (4-mm) been installed correctly? If it has not, install one as directed in Section 3.5, Installing the Anion Trap Column, and watch the background conductivity. If the background conductivity is now low, this means that the ATC is trapping contaminants from the eluent. The eluents probably have too many impurities (see items 1 - 3 above).

If the ATC is already installed, remove it. Is the background conductivity still high? If the background conductivity decreases, the ATC is the source of the high background conductivity.

A. Disconnect either the ATC-3 (2-mm) or the ATC-3 (4-mm) from the injection valve and direct the outlet to waste.

B. Flush the ATC with 200 mL of 70 mM $Na_2B_4O_7$. Use a flow rate of 0.5 mL/min on a 2-mm system or a flow rate of 2.0 mL/min on a 4-mm system.

C. Equilibrate the ATC with the strongest eluent used during the gradient run. Use a flow rate of 0.5 mL/min on a 2-mm system or a flow rate of 2.0 mL/min on a 4-mm system.

D. If the problem persists, replace the ATC.

6.2.3 A Contaminated Guard or Analytical Column

Remove the IonPac AG12A Guard and AS12A Analytical Columns from the system. If the background conductivity decreases, the column(s) is (are) the cause of the high background conductivity. Clean or replace the AG12A at the first sign of column performance degradation (compared to the original test chromatogram) to eliminate downtime. Clean the column(s) as instructed in “Column Care.”

6.2.4 Contaminated Hardware

To eliminate the hardware as the source of the high background conductivity, bypass the columns and the Anion Self-Regenerating Suppressor. Pump deionized water with a specific resistance of 18.2 megohm-cm through the system. The background conductivity should be less than 2 µS. If it is not, check the detector/conductivity cell calibration by injecting deionized water directly into it. See the appropriate manual for details.

6.2.5 A Contaminated ASRS-ULTRA Suppressor

This section describes routine cleanup procedures for the Anion Self-Regenerating Suppressors (ASRS-ULTRA) in the case of contamination. Consult the Troubleshooting Guide (see Section 4, “Troubleshooting Guide”) to first determine that the system

is operating properly. If the ASRS-ULTRA is determined to be the source of higher than normal back pressure, higher than anticipated conductivity, decreased suppression capacity or decreased sensitivity, cleaning the membrane may restore the performance of the system. Use the following procedures to clean the membrane.

Metal Contaminants or Precipitates

NOTE

The suppressor voltage is a good indicator of the resistance across the suppressor. Higher resistance may indicate contamination of the suppressor. For more information regarding monitoring the voltage, see Document No. 031814-02, "Removal of Iron Contamination from Electrolytic Suppressors."

A. Turn off the SRS Control unit.
B. Disconnect the analytical (and guard) column(s) from the injection valve and the ASRS-ULTRA. Refer to the specific analytical column Product Manual for column cleanup procedures.

C. If you are running in the **AutoSuppression External Water Mode**, turn off the external water and disconnect the external water line from the ASRS-ULTRA **REGEN IN** port.

D. Disconnect the liquid line from the ASRS-ULTRA **ELUENT OUT** port to the cell at the cell fitting and reconnect it to the **REGEN IN** port.

E. Connect a temporary line from the priming block or the low-pressure tee on the isocratic or gradient pump to a container with a solution of 0.2 M oxalic acid. Pump this solution through the ASRS-ULTRA (4-mm) at 1-2 mL/min for 30 minutes. For 2-mm systems pump this solution through the ASRS-ULTRA (2-mm) at 0.25-0.50 mL/min for 30 minutes.

NOTE

Bypassing internal pump manifolds when temporarily pumping high concentration cleaning solutions significantly reduces the time required to reequilibrate the system to low concentration eluents.

F. Flush the ASRS-ULTRA with deionized water for 10 minutes.

G. Perform steps A - D of the procedure in Section 4.1, "Small Analyte Peak Areas."

H. Turn on the SRS Control unit for the **AutoSuppression Recycle or External Water Modes** of operation. Ensure that the SRS Control unit is **off** for the **Chemical Suppression Mode** of operation.

I. Flush the ASRS-ULTRA with eluent for 10 minutes.

J. Reinstall the analytical (and guard) column(s). Begin pumping eluent through the system at the flow rate required for your analysis and equilibrate the system.

6.2.6 A Contaminated AMMS III Suppressor

A. Check the regenerant flow rate at the REGEN OUT port of the AMMS. For the example isocratic applications, this flow rate should be 3 - 5 mL/min.

B. Check the eluent flow rate. In general, the eluent flow rate for 4-mm applications, it should be 1.0 mL/min. Refer to the Anion MicroMembrane Suppressor Product Manual (Document No. 034449-02) for assistance in determining that the eluent is within suppressible limits.

C. If you are using an AutoRegen Accessory with the SRS (in the Chemical Suppression Mode) or the MMS, prepare fresh regenerant solution. Test both the suppressor and the AutoRegen Regenerant Cartridge for contamination.

1. **If the background conductivity is high after preparing fresh regenerant and bypassing the AutoRegen Regenerant Cartridge, you probably need to clean or replace your SRS or MMS.**

2. If the background conductivity is low when freshly prepared regenerant is run through the SRS or MMS without an AutoRegen Accessory in-line, test the AutoRegen Regenerant Cartridge to see if it is **expended.** Connect the freshly prepared regenerant to the AutoRegen Regenerant Cartridge. Pump approximately 200 mL of regenerant through the AutoRegen Regenerant Cartridge to waste before recycling the regenerant back to the regenerant reservoir. If the background conductivity is high after placing the AutoRegen Accessory in-line, you probably need to replace the AutoRegen Regenerant Cartridge. Refer to the "AutoRegen Regenerant Cartridge Refill Product Manual" (Document No. 032852) for assistance.

6.2.7 A Contaminated Anion Atlas Electrolytic Suppressor, AAES

Metal Contaminants or Precipitates

A. Turn off the power to the AAES.

B. Disconnect the analytical (and guard) column(s) from the injection valve and the AAES. Refer to the specific analytical column Product Manual for column cleanup procedures.

C. If you are running in the **AutoSuppression External Water Mode**, turn off the external water and disconnect the external water line from the AAES **REGEN IN** port.

D. Disconnect the liquid line from the AAES **ELUENT OUT** port to the cell at the cell fitting and reconnect it to the **REGEN IN** port.

E. Connect a temporary line from the priming block or the low-pressure tee on the isocratic or gradient pump to a container with a solution of 0.5 M oxalic acid. Pass 60 mL of this solution through the AAES using the Trap Column / Suppressor Clean-up Kit (P/N 059649) or pump this solution through the AAES at 2.0 mL/min for 30 minutes.

NOTE

Bypassing internal pump manifolds when temporarily pumping high concentration cleaning solutions significantly reduces the time required to re-equilibrate the system to low concentration eluents.

F. Flush the AAES with deionized water at 2 mL/min for 30 minutes.

G. Reinstall the AAES according to procedures in Section 4.2.1, "Eluent and Regenerant Flow Path Connections in the AutoSuppression Recycle Mode Operation" or Section 4.3.1, "Eluent Flow Path Connections in the AutoSuppression External Water Mode Operation" and resume operation.

Organic Contaminants

A. Turn off the power to the AAES.

B. Disconnect the analytical (and guard) column(s) from the injection valve and the AAES. Refer to the specific analytical column Product Manual for column cleanup procedures.

C. If you are running in the **AutoSuppression External Water Mode**, turn off the external water and disconnect the external water line from the AAES REGEN IN port. If you are running in the **AutoSuppression Recycle Mode**, proceed to D.

D. Disconnect the liquid line from the AAES **ELUENT OUT** port to the cell at the cell fitting and reconnect it to the **REGEN IN** port.

E. Connect a temporary line from the priming block or the low-pressure tee on the isocratic or gradient pump to a

container with a solution of freshly prepared 10% 1.0 M H_2SO_4/90% acetonitrile. H_2SO_4/acetonitrile solutions are not stable during long term storage so this cleanup solution must be made immediately before each column cleanup. Alternatively, it can be proportioned from 1 bottle containing 1.0 M H_2SO_4 and another bottle containing 100% acetonitrile. Pass 60 mL of this solution through the AAES using the Trap Column / Suppressor Clean-up Kit (P/N 059649) or pump this solution through the AAES at 1.0 mL/min for 60 minutes.

NOTE

Bypassing internal pump manifolds when temporarily pumping high concentration cleaning solutions significantly reduces the time required to re-equilibrate the system to low concentration eluents.

F. Flush the AAES with deionized water at 2 mL/min for 30 minutes.

6.3 Poor Peak Resolution

Poor peak resolution can be due any or all of the following factors.

6.3.1 Loss of Column Efficiency

A. Check to see if headspace has developed in the guard or analytical column. This is usually due to improper use of the column such as submitting it to high pressures. Remove the column's top end fitting (see Section 6.1.2, "Replacing Column Bed Support Assemblies"). If the resin does not fill the column body all the way to the top, it means that the resin bed has collapsed, creating a headspace. The column must be replaced.

B. Extra-column effects can result in sample band dispersion, making the peaks' elution less efficient. Make sure you are using PEEK tubing with an ID of no greater than 0.010" for 4-mm systems or no greater than 0.005" for 2-mm systems to make all eluent liquid line connections between the injection valve and the detector cell inlet. Cut the tubing lengths as short as possible. Check for leaks.

6.3.2 Poor Resolution Due to Shortened Retention Times

Even with adequate system and column efficiency, resolution of peaks will be compromised if analytes elute too fast.

A. Check the flow rate. See if the eluent flow rate is equivalent to the flow rate specified by the analytical protocol. Measure the eluent flow rate after the column using a stopwatch and graduated cylinder.

B. Check to see if the eluent compositions and concentrations are correct. An eluent that is too concentrated will cause the peaks to elute faster. Prepare fresh eluent. If you are using a gradient pump to proportion the eluent, components from two or three different eluent reservoirs, the resulting eluent composition may not be accurate enough for the application. Use one reservoir containing the correct eluent composition to see if this is the problem. This may be a problem when one of the proportioned eluents is less than 5%.

C. Column contamination can lead to a loss of column capacity. This is because all of the anion exchange sites will no longer be available for the sample ions. For example, polyvalent anions from the sample or metals may concentrate on the column. Refer to "Column Care," for recommended column cleanup procedures.

Possible sources of column contamination are impurities in chemicals and in the deionized water used for eluents or components of the sample matrix. Be especially careful to make sure that the recommended chemicals are used. The deionized water should have a specific resistance of 18.2 megohm-cm.

D. Diluting the eluent will improve peak resolution, but will also increase the analytes' retention times. If a 10% dilution of the eluent is not sufficient to obtain the desired peak resolution, or if the resulting increase in retention times is unacceptable, clean the column (see "Column Care").

After cleaning the column, reinstall it in the system and let it equilibrate with eluent for about 30 minutes. No water wash is necessary. The column is equilibrated when consecutive injections of the standard give reproducible retention times. The original column capacity should be restored by this treatment, since the contaminants should be eluted from the column. If you need assistance in solving resolution problems, contact the nearest DIONEX Office (see, "DIONEX Worldwide Offices")

6.3.3 Loss of Front End Resolution

If poor resolution or efficiency is observed for the peaks eluting near the system void volume compared to the later eluting peaks, check the following:

A. Improper eluent concentration may be the problem. Remake the eluent as required for your application. Ensure that the water and chemicals used are of the required purity.

B. Column overloading may be the problem. Reduce the amount of sample ions being injected onto the analytical column by either diluting the sample or injecting a smaller volume onto the column.

C. Sluggish operation of the injection valve may be the problem. Check the air pressure and make sure there are no gas leaks or partially plugged port faces. Refer to the valve manual for instructions.

D. Improperly swept out volumes anywhere in the system prior to the guard and analytical columns may be the problem. Swap components, one at a time, in the system prior to the analytical column and test for front-end resolution after every system change.

6.3.4 Spurious Peaks

A. The columns may be contaminated. If the samples contain an appreciable level of polyvalent ions and the column is used with a weak eluent system, the retention times for the analytes will then decrease and be spurious, inefficient (broad) peaks that can show up at unexpected times. Clean the column as indicated in "Column Care."

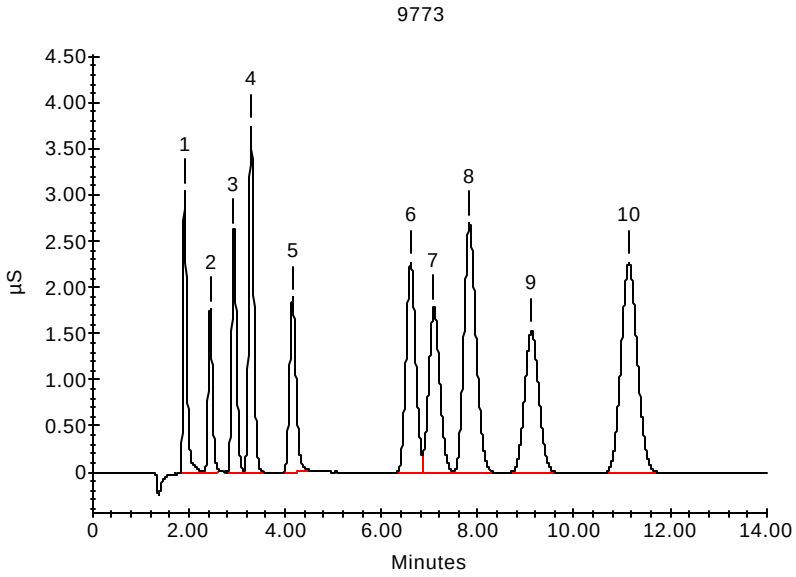
If you need assistance in determining the best way to clean strongly retained solutes in your specific sample matrix from the IonPac AS12A columns, contact the nearest DIONEX Office (see, "DIONEX Worldwide Offices").

B. The injection valve may need maintenance. When an injection valve is actuated, the possibility of creating a baseline disturbance exists. This baseline upset can show up as a peak of varying size and shape. This will occur when the injection valve needs to be cleaned or retorqued (see valve manual). Check to see that there are no restrictions in the tubing connected to the valve. Also check the valve port faces for blockage and replace them if necessary. Refer to the Valve Manual for troubleshooting and service procedures. Small baseline disturbances at the beginning or at the end of the chromatogram can be overlooked as long as they do not interfere with the quantification of the peaks of interest.

For DX-300 systems equipped with a Rheodyne Microinjection Valve, Model 9126 (DIONEX P/N 044697), consult the accompanying manual for service instructions.

IonPac® AS12A
Analytical (4 x 200 mm)
Product No. 46034

Serial No. : 9773 **Pressure (PSI) : 1650** **Date : 5/18/00 9:46:33 AM**



Eluent: 2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Flow Rate: 1.5 mL/min
Detection: Suppressed Conductivity
ASRS®-ULTRA
AutoSuppression® Recycle Mode

Injection Volume: 10 µL

Storage Solution: 0.1 M NaOH

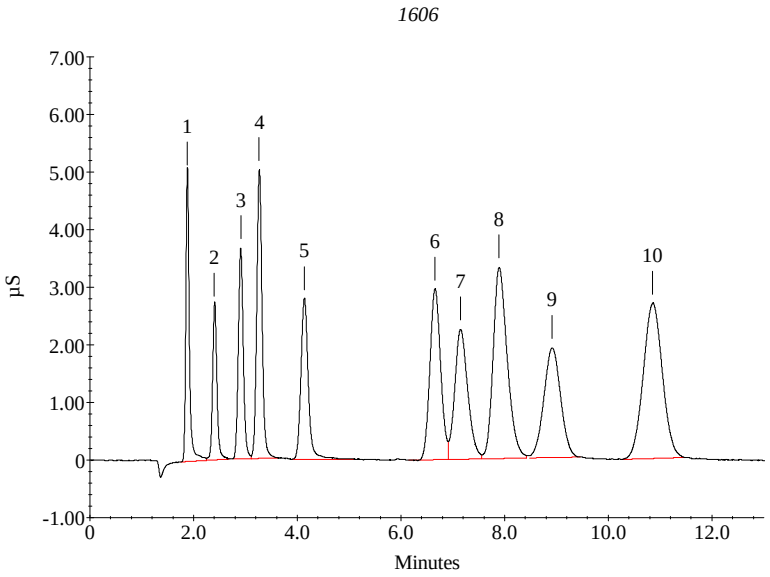
Peak Information : Found Components

Peak No.	Retention Time	Name	(mg/L)	Efficiency	Asymmetry (10%)	Resolution
1	1.91	Fluoride	3.0	3158	1.8	3.61
2	2.44	Chlorite	10.0	3786	1.5	2.84
3	2.93	Bromate	20.0	3931	1.5	1.87
4	3.29	Chloride	6.0	4406	1.3	3.81
5	4.15	Nitrite	10.0	4241	1.3	7.85
6	6.61	Bromide	20.0	5001	1.2	1.21
7	7.09	Chlorate	20.0	4456	n/a	1.67
8	7.83	Nitrate	20.0	4546	1.3	2.54
9	9.12	Phosphate	30.0	4318	1.1	3.42
10	11.14	Sulfate	20.0	5022	1.2	n/a

File Name : C:\PEAKNET\DATA\EXAMPLES\46034 AS12A 4MM_A015.DXD

IonPac® AS12A
Analytical (2 x 200 mm)
Product No. 46055

Serial No. : 1606 **Pressure (PSI) : 1560** **Date : 4/24/00 10:11:26 AM**



Eluent: 2.7 mM Na_2CO_3/0.3 mM $NaHCO_3$
Flow Rate: 0.38 mL/min
Detection: Suppressed Conductivity at 10µSFS
ASRS®-ULTRA, 2-mm
AutoSuppression® Recycle Mode

Injection Volume: 2.5 µL

Storage Solution: 0.1 M NaOH

Peak Information : Found Components

Peak No.	Retention Time	Name	(mg/L)	Efficiency	Asymmetry (10%)	Resolution
1	1.88	Fluoride	3.0	4382	1.7	4.04
2	2.40	Chlorite	10.0	4248	1.4	3.18
3	2.91	Bromate	20.0	4534	1.2	2.01
4	3.27	Chloride	6.0	5037	1.2	4.01
5	4.13	Nitrite	10.0	4400	1.2	7.95
6	6.65	Bromide	20.0	4768	n/a	1.19
7	7.15	Chlorate	20.0	3933	n/a	1.57
8	7.89	Nitrate	20.0	4150	1.5	1.88
9	8.92	Phosphate	30.0	3528	1.0	3.04
10	10.86	Sulfate	20.0	4082	1.1	6.14

File Name : I:\LBURHANUDIN\QA REPORT BOOK _PDF_DXD_FILES\46055_AS12A-2MM_A001.DXD

RECOMMENDED OPERATION PRESSURES

Operating a column above its recommended pressure limit can cause irreversible loss of column performance. The maximum recommended operating pressure for IonPac AS12A columns is 3,500 psi (24.13 MPa).

CAUTION

Do not run deionized water through the column for longer than 15 minutes at 1 mL/min (4-mm systems) or 0.25 mL/min (2-mm systems)

COLUMN START-UP

The column is shipped using the eluent as the storage solution.

Prepare the eluent shown on the test chromatogram, install the column in the chromatography module and test the column performance under the conditions described in the test chromatogram. Continue making injections of the test standard until consecutive injections of the standard give reproducible retention times. Equilibration is complete when consecutive injections of the standard give reproducible retention times.

COLUMN STORAGE

For both short-term and long-term storage, use eluent for the column storage solution. Flush the column for a minimum of 10 minutes with the storage solution (eluent). Cap both ends securely, using the plugs supplied with the column.

COLUMN CLEANUP

The following column cleanup protocols have been divided into three general isocratic protocols to remove acid-soluble, base-soluble or organic contaminants. They can be combined into one gradient protocol if desired but the following precautions should be observed.

Always ensure that the cleanup protocol used does not switch between eluents which may create high pressure eluent interface zones in the column. High pressure zones can disrupt the uniformity of the packing of the column bed and irreversibly damage the performance of the column. High pressure zones in the column can be created by pumping successive eluents through the column that are not miscible, that have eluent components in one eluent that will precipitate out in the other eluent or by using an acid eluent followed by a base eluent which may create a neutralization pressure band. The precipitation of the salts in solvents during column rinses can result in very high pressure zones. High viscosity mixing zones can be created between two eluents having solvents with a very high energy of mixing.

When in doubt, always include short column rinse steps to reduce the solvent content of the eluent to $\leq$ 5% levels and the ionic strength of the eluent to $\leq$ 50 mM levels to avoid creating high pressure zones in the column that may disrupt the uniformity of the column packing.

CHOOSING THE APPROPRIATE CLEANUP SOLUTION

A. Hydrophilic ionic contamination of low valency

1. 10X eluent concentrate
   (27 mM Na_2CO_3/3 mM $NaHCO_3$ = 10X concentrate of the production test eluent)

B. High valency hydrophobic ions

1. 200 mM HCl in 80% acetonitrile
   The acetonitrile solution is stored in a separate eluent bottle because acetonitrile slowly breaks down in acidic aqueous solutions. Prepare two bottles (E1 and E2) with the following 500 mL solutions:

E1: 100% acetonitrile
E2: 1 M HCl using degassed Type I Reagent Grade Water with a specific resistance of 18.2 megohm-cm.

You may make 200 mM HCl in 80% acetonitrile in a single bottle but this solution must be made immediately before each use and cannot be stored because of acetonitrile degradation

C. Metal contamination

1. 0.1 M oxalic acid.
   Iron or aluminum contamination often results in tailing of sulfate and phosphate. Aluminum contamination can also result in low phosphate recoveries.

D. Regardless of the cleanup solution chosen, use the following cleanup procedure, "Column Cleanup Procedure," to clean the AG12A and AS12A.

COLUMN CLEANUP PROCEDURE

A. **Prepare a 500 mL solution of the appropriate cleanup solution** using the guidelines, "Choosing the Appropriate Cleanup Solution," above.

B. **Disconnect the ASRS-ULTRA or AMMS** from the IonPac AS12A Analytical Column. If your system is configured with both a guard column and an analytical column, reverse the order of the guard and analytical column in the eluent flow path. Double check that the eluent flows in the direction designated on each of the column labels.

CAUTION

When cleaning an analytical column and a guard column in series, ensure that the guard column is placed after the analytical column in the eluent flow path. Contaminants that have accumulated on the guard column can be eluted onto the analytical column and irreversibly damage it. If in doubt, clean each column separately.

C. **Set the pump flow rate to 1.0 mL/min for an AS12A 4-mm Analytical or Guard Column, or set the pump flow rate to 0.25 mL/min for an AS12A 2-mm Analytical or Guard Column.**

D. **Rinse the column for 15 minutes with deionized water** before pumping the chosen cleanup solution over the column.

E. **Pump the cleanup solution through the column for at least 60 minutes.**

F. **Rinse the column for 15 minutes with deionized water** before pumping eluent over the column.

G. **Equilibrate the column(s) with eluent** before resuming normal operation for at least 30 minutes.

H. **Reconnect the ASRS-ULTRA or AMMS** to the AS12A Analytical Column and **place the guard column in line** between the injection valve and the analytical column if your system was originally configured with a guard column.