

Errata

Product Manual for Dionex IonPac™ AS11-HC and AG11-HC Columns
031333-09

For new orders of the following parts discussed in this manual, please use the updated part numbers listed below.

| Part | Old Part Number in this manual | Updated Part Number to use for new orders |
|-------------------------------------|---------------------------------------|--|
| <i>PROD,COL,IP,ATC-3,4X35MM</i> | <i>059661</i> | <i>079932</i> |
| <i>PROD,COL,IP,UTAC-LP2,4X35MM</i> | <i>072779</i> | <i>079917</i> |
| <i>PROD,COL,IP,UTAC-ULP2,5X23MM</i> | <i>072780</i> | <i>079918</i> |



Thermo Scientific

IonPac™ AS11-HC

Product Manual

P/N: 031333-09 February 2012

Product Manual

for

Thermo Scientific Dionex IonPac AG11-HC Guard Column

(4 x 50 mm, P/N 052962)

(2 x 50 mm, P/N 052963)

Thermo Scientific Dionex IonPac AG11-HC Capillary Guard Column

(0.4 x 50 mm, P/N 078430)

Thermo Scientific Dionex IonPac AS11-HC Analytical Column

(4 x 250 mm, P/N 052960)

(2 x 250 mm, P/N 052961)

Thermo Scientific Dionex IonPac AS11-HC Capillary Column

(0.4 x 250 mm, P/N 078429)

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SECTION 1 – INTRODUCTION

The Dionex IonPac AS11-HC Columns are specifically designed to resolve a large number of inorganic anions and organic acid anions from a single sample injection in one gradient run using hydroxide eluent systems.

The Dionex IonPac AS11-HC is a high capacity column with selectivity similar to the Thermo Scientific Dionex AS11. The high capacity allows injection of more concentrated samples without overloading and peak broadening. Hydroxide is normally used for gradient elution to minimize background shift. Because of high background conductance, sodium carbonate/bicarbonate eluents are not appropriate for gradient analysis but can be used for isocratic applications. By using a hydroxide gradient, strongly retained trivalent ions, such as phosphate and citrate, are efficiently eluted in the same run that also gives baseline resolution of the weakly retained monovalent anions; fluoride, lactate, acetate, formate, and butyrate.

Another benefit of using the Dionex IonPac AS11-HC column is the ability to easily change the order of elution of ions with different valencies simply by changing the gradient profile. For example, if citrate is present in high enough concentration to interfere with chromate, the chromate peak can be moved ahead of the citrate peak by using a slightly different gradient. Dionex IonPac AS11-HC columns are available in 0.4 x 250 mm, 2 x 250 mm and 4 x 250 mm formats, thus supporting flow rates from 0.015 mL/min to 3.0 mL/min. Dionex IonPac AS11-HC columns are stable between pH 0 and 14 and are compatible with eluents containing 0-100% organic solvents. The Dionex IonPac AG11-HC guard column is packed with a microporous resin with a lower capacity. The microporous resin ensures optimum long term performance of the guard column.

The Dionex IonPac AS11-HC Capillary Column (0.4 x 250 mm) is packed with the same material as the equivalent standard bore version (producing the same performance as a 4 mm column), but requires only one-hundredth (1/100) the eluent flow rate. The capillary format has the advantage of less eluent consumption, providing reduced costs.

Table 1
Dionex IonPac AS11-HC/Dionex IonPac AG11-HC Packing Specifications

| Column | Particle Diameter μm | Substrate X-linking % | Latex Diameter nm | Column Capacity $\mu\text{eq/column}$ | Functional Group | Hydrophobicity |
|---------------------------|---------------------------------|-----------------------|-------------------|---------------------------------------|-----------------------------|----------------|
| AS11-HC* 4x250 mm | 9 | 55% | 70 | 290 | Alkanol quaternary ammonium | Medium-Low |
| AG11-HC ** 4x50 mm | 13 | 55% | 70 | 7 | Alkanol quaternary ammonium | Medium-Low |
| AS11-HC * 2x250 mm | 9 | 55% | 70 | 72.5 | Alkanol quaternary ammonium | Medium-Low |
| AG11-HC ** 2x50mm | 13 | 55% | 70 | 1.75 | Alkanol quaternary ammonium | Medium-Low |
| AS11-HC * 0.4 x 250 mm | 9 | 55% | 70 | 2.90 | Alkanol quaternary ammonium | Medium-Low |
| AG11-HC ** 0.4 x 50 mm | 13 | 55% | 70 | 0.07 | Alkanol quaternary ammonium | Medium-Low |

* Analytical/Capillary Column resin composition: supermacroporous polyvinylbenzyl ammonium polymer cross-linked with divinylbenzene.

** Guard Column resin composition: microporous polyvinylbenzyl

Table 2
Dionex IonPac AS11-HC/Dionex IonPac AG11-HC Operating Parameters

| Column | Typical Back Pressure psi (MPa) | Standard Flow Rate mL/min | Maximum Flow Rate mL/min |
|---|------------------------------------|---------------------------------|-----------------------------|
| AS11-HC 4-mm Analytical | ≤ 1,900 (13.10) | 1.50 | 3.00 |
| AG11-HC 4-mm Guard | ≤ 150 (1.03) | 1.50 | 3.00 |
| AS11-HC and AG11-HC 4-mm columns | ≤ 2,050 (14.13) | 1.50 | 3.00 |
| AS11-HC 2-mm Analytical | ≤ 1,800 (12.41) | 0.38 | 0.75 |
| AG11-HC 2-mm Guard | ≤ 150 (1.03) | 0.38 | 0.75 |
| AS11-HC and AG11-HC 2-mm columns | ≤ 1,950 (13.44) | 0.38 | 0.75 |
| AS11-HC 0.4-mm Capillary | ≤ 2,000 (13.79) | 0.015 | 0.03 |
| AG11-HC 0.4-mm Capillary Guard | ≤ 200 (1.38) | 0.015 | 0.03 |
| AS11-HC and AG11-HC 0.4-mm Capillary and Capillary Guard | ≤ 2200 (15.17) | 0.015 | 0.03 |



For assistance, contact Technical Support for Dionex Products. In the U.S., call 1-800-346-6390. Outside the U.S., call the nearest Thermo Fisher Scientific office.

NOTE

SECTION 2 – ION CHROMATOGRAPHY SYSTEMS

The proper configuration of an Ion Chromatography System (ICS) in 2-mm or 4-mm format is based on the ratio of the 2-mm to 4-mm column cross-sectional area (a factor of 1/4). The selected format will affect the type of pump recommended. A gradient pump is designed to blend and pump isocratic, linear, or gradient mixtures of up to four mobile phase components at precisely controlled flow rates. An isocratic pump is for applications not requiring gradient and multi-eluent proportioning capabilities. Both are offered in either standard bore or microbore options.

- For an ICS in 2-mm format, a microbore isocratic pump, standard bore isocratic pump, microbore gradient pump, or standard bore gradient pump is recommended.
- For an ICS in 4-mm format, a standard bore isocratic pump or standard bore gradient pump is recommended.
- For an ICS in 0.4 mm format, a Capillary IC system such as the ICS-5000 system is recommended.

See Appendix B, "System Configuration" for specific recommended settings and parts including pumps, eluent flow rate, Thermo Scientific Dionex Self-Regenerating Suppressor (SRS), Thermo Scientific Dionex MicroMembrane Suppressor (MMS), Thermo Scientific Dionex Capillary Electrolytic Suppressor (CES), injection loop, system void volume, detectors, and tubing back pressure.



NOTE

Do not operate suppressors over 40 °C. If application requires a higher temperature, place the suppressor outside of the chromatographic oven. Use of a Dionex EG with a Dionex EGC III KOH (P/N 074532) cartridge for gradient applications is highly recommended for minimum baseline change when performing eluent step changes or gradients.

SECTION 3 – INSTALLATION

3.1. System Requirements

3.1.1. System Requirements for 0.4mm Operation

The Dionex IonPac AS11-HC 0.4 mm Capillary Guard and Capillary Column are designed to be run on a capillary ion chromatograph system equipped with suppressed conductivity detection. It is recommended to run the capillary column only on the Dionex ICS-5000 capillary system for best performance.

3.1.2. System Requirements for 2-mm Operation

The Dionex IonPac AS11-HC 2-mm Guard and Analytical Columns are designed to run on 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 pump with standard (1/8" pistons) pump heads. For isocratic analyses at flow rates below 0.5 mL/min and gradient analyses, a microbore pump (1/16" pistons) must be employed.

3.1.3. System Requirements for 4-mm Operation

The Dionex IonPac AS11-HC 4-mm Guard and Analytical Columns are designed to 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 pump with a standard pump heads (1/8" pistons). Isocratic analysis can also be performed on a pump with standard bore pump heads (1/8" pistons).

3.1.4. 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) i.d. PEEK tubing. 0.010" i.d. 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.

3.2. The Sample Concentrator

The Dionex Trace Anion Concentrator Low Pressure Column (Dionex TAC-LP1, P/N 046026), the Dionex Trace Anion Concentrator Ultra Low Pressure Column (Dionex TAC-ULP1, P/N 061400), the Dionex Ultra Trace Anion Concentrator Low Pressure Column (Dionex UTAC-LP1, P/N 063079) or (Dionex UTACLP2, P/N 072779), the Dionex Ultra Trace Anion Concentrator Ultra Low Pressure Column (Dionex UTAC-ULP1, P/N 063475) or (Dionex UTAC-ULP2, P/N 072780), the Dionex Ultra Trace Anion Concentrator Extremely Low Pressure Column (Dionex UTAC-XLP1, P/N 063459) or (Dionex UTAC-XLP2, P/N 072781), or the Dionex IonPac AG11-HC Guard Column can be used for trace anion concentration work with the 2-mm and 4-mm Dionex IonPac AS11-HC columns. For trace anion concentration work with the 0.4-mm Dionex IonPac AS11-HC column, use the Dionex IonSwift MAC-100 Concentrator Column. 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 and a detailed discussion of anion concentration techniques refer to:

- Section 3, “Operation,” of the Dionex Trace Anion Concentrator Low Pressure (Dionex TAC-LP1) and Dionex Ultra Low Pressure (Dionex TAC-ULP1) Column Product Manual (Document No. 034972).
- Section 3, “Operation,” of the Dionex Ultra Trace Anion Concentrator Low Pressure (Dionex UTAC-LP1), Dionex Ultra Low Pressure (Dionex UTAC-ULP1), and Dionex Extremely Low Pressure (Dionex UTAC-XLP1) Column Product Manual (Document No. 065091.)
- Section 4, “Operation,” of the Dionex Ultra Trace Anion Concentrator 2 Low Pressure (Dionex UTAC-LP2), Dionex Ultra Low Pressure (Dionex UTAC-ULP2), and Dionex Extremely Low Pressure (Dionex UTAC-XLP2) Column Product Manual (Document No. 065376.)

**CAUTION**

Dionex IonPac Trace Anion Concentrator Column, Dionex TAC-2 (P/N 043101), is not optimized for use with hydroxide eluents and should not be used for concentrator work with the Dionex IonPac AS11-HC. Instead, Concentrators (Dionex TAC-LP1, TAC-ULP1, UTAC 1, UTAC 2 or Dionex IonSwift MAC-100) or Guards (Dionex IonPac AG11-HC 4 mm or Dionex IonPac AG11-HC 2 mm) should be used.

3.3. The Injection Loop

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. Generally, you should not inject more than 10 nanomoles of any one analyte onto a 2-mm analytical column. Injecting larger number of moles of a sample can result in overloading the column which can affect the detection linearity. For low concentrations of analytes, larger injection loops can be used to increase sensitivity. The Dionex IonPac AS11-HC 2-mm requires a microbore HPLC system configuration. Install an injection loop one-fourth or less ($<15 \mu\text{L}$) of the loop volume used with a 4-mm analytical system.

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 is sufficient. Generally, you should not inject more than 40 nanomoles of any one analyte onto the 4-mm analytical column. Injecting larger number of moles of a sample can result in overloading the column which can affect the detection linearity. For low concentrations of analytes, larger injection loops can be used to increase sensitivity.

3.3.3. The 0.4 mm System Injection Loop, 0.4 μL Internal Loop

For most applications on a 0.4 mm capillary system, a 0.4 μL injection loop is sufficient. Generally, do not inject more than 0.5 nanomoles of any one analyte into a 0.4 mm capillary column. Injecting larger numbers of moles of a sample can result in overloading the column, which can affect the detection linearity. For samples containing low concentrations of analytes, larger injection loops can be used to increase sensitivity.

3.4. The Dionex IonPac AG11-HC Guard Column

A Dionex IonPac AG11-HC Guard Column is normally used with the Dionex IonPac AS11-HC Analytical/Capillary Column. Retention times will increase by approximately 2% when a guard column is placed in-line prior to the analytical/capillary column. A guard is placed prior to the analytical/capillary column to prevent sample contaminants from eluting onto the analytical/capillary column. It is easier to clean or replace a guard column than it is an analytical/capillary column. Replacing the Dionex IonPac AG11-HC Guard Column at the first sign of peak efficiency loss or decreased retention time will prolong the life of the Dionex IonPac AS11-HC Analytical/Capillary Column.

3.5. Installing the Dionex CR-ATC Trap Column for Use with Dionex EGC

For Dionex IonPac AS11-HC applications using the Dionex EGC KOH cartridge, a Dionex CR-ATC Continuously Regenerated Trap Column (P/N 060477 or 072078) should be installed at the Dionex EGC eluent outlet to remove trace level anionic contaminants from the carrier deionized water. See the Dionex CR-TC Product Manual (Document No. 031910) for instructions. As an alternative for 2 mm and 4 mm columns, the Dionex ATC-HC Trap Column (P/N 059604) should be installed between the pump outlet and the inlet of the Dionex EluGen Cartridge to remove anionic contaminants from the carrier deionized water. See the Dionex ATC-HC Product Manual (Document No. 032697) for instructions.

If the lower capacity Dionex ATC-3 Trap Column (P/N 059660 and 059661) is used with 2 mm and 4 mm columns, it should be installed between the gradient pump and the injection valve to remove anionic contaminants from the eluent. The Dionex ATC-3 column is used when performing sodium hydroxide gradient anion exchange applications using hand-prepared bottled eluents. See the Dionex ATC-3 Product Manual (Document No. 032697) for instructions.

The Dionex ATC-HC (P/N 059604) and Dionex ATC-3 Trap Columns will require off-line regeneration. To use the Dionex ATC-HC or Dionex ATC-3 Anion Trap Columns, refer to the Product Manuals.

3.6. Eluent Storage

Dionex IonPac AS11-HC columns are designed to be used with hydroxide 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. Dionex Anion Self-Regenerating Suppressor Requirements

A Dionex 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 Dionex ASRS 300 modes of operation.



CAUTION

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

For Dionex IonPac AS11-HC 4-mm Analytical Column, use a Dionex ASRS 300 (4-mm, P/N 061561).

For Dionex IonPac AS11-HC 2-mm Analytical Column, use a Dionex ASRS 300 (2-mm, P/N 061562).

For Dionex IonPac AS11-HC 0.4-mm Capillary Column, use a Dionex ACES 300 (2-mm, P/N 072052).

For detailed information on the operation of the Dionex Anion Self-Regenerating Suppressor, see Document No. 031956, the "Product Manual for the Dionex Anion Self-Regenerating Suppressor 300, the Dionex ASRS 300."

For detailed information on the operation of the Dionex Anion Capillary Electrolytic Suppressor 300, see Document No. 065386, the "Product Manual for the Dionex Anion Capillary Electrolytic Suppressor 300, the Dionex ACES 300"

3.8. Dionex Anion MicroMembrane Suppressor Requirements

A Dionex Anion MicroMembrane Suppressor (Dionex AMMS 300) may be used instead of a Dionex ASRS 300 for applications that require suppressed conductivity detection. Use a Dionex AMMS 300 (P/N 064558) with the Dionex IonPac AS11-HC 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 Dionex AMMS 300 (2-mm) (P/N 064559).

For detailed information on the operation of the Dionex Anion MicroMembrane Suppressor, see Document No. 031727, the "Product Manual for the Dionex Anion MicroMembrane Suppressor 300, the Dionex AMMS 300".

3.9. Using Displacement Chemical Regeneration (DCR) with the Chemical Suppression Mode

The Dionex Displacement Chemical Regeneration (Dionex DCR) Mode is recommended for chemical suppression using sulfuric acid and the Dionex Anion MicroMembrane Suppressor (Dionex AMMS 300). See the DCR kit manual, Document P/N 031664, for details.



Use proper safety precautions in handling acids and bases.

SAFETY

3.10. Using the Dionex EGC-KOH with Dionex IonPac AS11-HC

The Dionex IonPac AS11-HC column is recommended for use with Thermo Scientific Dionex ICS-2100, or Dionex ICS-5000 IC Systems equipped with a Dionex Eluent Generator. The Dionex Eluent Generator is used to automatically produce potassium hydroxide gradients from deionized water. The Dionex IonPac AS11-HC can be used with older Dionex IC Systems equipped with an Eluent Generator or a Dionex RFC-30 Reagent Free Controller. Please refer to the Dionex EG40 manual, Document No. 031373, for information on the operation of the Dionex EG40. Please refer to the Dionex EG50 Product Manual, Document No. 031908, for information on the operation of the Dionex EG50.

3.10.1. Detector Requirements

See Section 2, “Ion Chromatography Systems,” for 2-mm, 4-mm and 0.4 mm system detector, cell and thermal stabilizer requirements.

3.11. Installation of the Capillary Column

1. Before installing the new separator column, cut off the column label and slide it into the holder on the front of the cartridge (see Figure 6).
2. For reference, Figure 1 shows the column cartridge after installation of both a capillary guard column and a capillary separator column. Figure 2 shows the column cartridge after installation of only a capillary separator column.

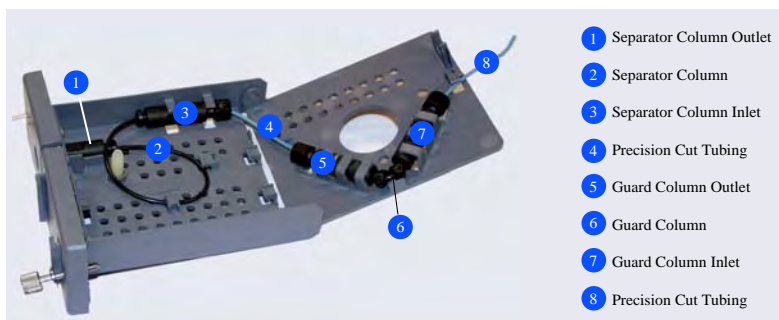


Figure 1
Separator and Guard Columns Installed in Column Cartridge

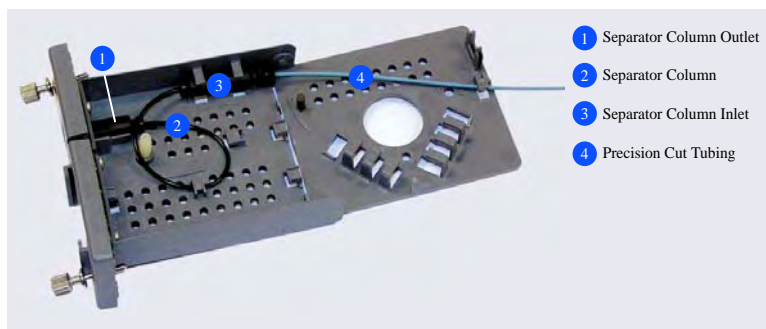


Figure 2
Separator Column Only Installed in Column Cartridge

3. Locate the Dionex IC Cube Tubing Kit (P/N 072186) that is shipped with the Dionex IC Cube. The tubing kit includes the following items:

Table 3
Contents of the Dionex IC Cube Tubing Kit (P/N 072186)

| Part | Length / Quantity | Part Number | Used To Connect |
|---|-------------------|-------------|--|
| Precision cut 0.062 mm (0.0025-in) ID PEEK tubing, blue | 65 mm (2.56 in) | 072188 | 50 mm guard column outlet to 250 mm separator column inlet |
| Precision cut 0.062 mm (0.0025-in) ID PEEK tubing, blue, labeled VALVE PORT 3 | 115 mm (4.53 in) | 072189 | Guard column inlet to injection valve |
| Precision cut 0.062 mm (0.0025-in) ID PEEK tubing, blue | 75 mm (2.93 in) | 074603 | 35 mm guard column outlet to 150 mm separator column inlet |
| Precision cut 0.062 mm (0.0025-in) ID PEEK tubing, blue, labeled VALVE PORT 3 | 210 mm (8.27 in) | 072187 | Separator column inlet to injection valve (if a guard column is not present) |
| 0.25 mm (0.010-in) ID PEEK tubing, black | 610 mm (24 in) | 042690 | EG degas cartridge REGEN OUT to waste (if an EG is not present) |
| Fitting bolt, 10-32 hex double-cone (smaller), black | 3 | 072949 | Connect precision cut 0.062 mm (0.0025-in) ID PEEK tubing |
| Fitting bolt, 10-32 double-cone (larger), black | 1 | 043275 | Connect 0.25 mm (0.010-in) ID PEEK tubing (black) |
| Ferrule fitting, 10-32 double-cone, tan | 4 | 043276 | Use with both sizes of fitting bolts |

4. Refer to the following figures for the precision cut tubing required for your configuration:

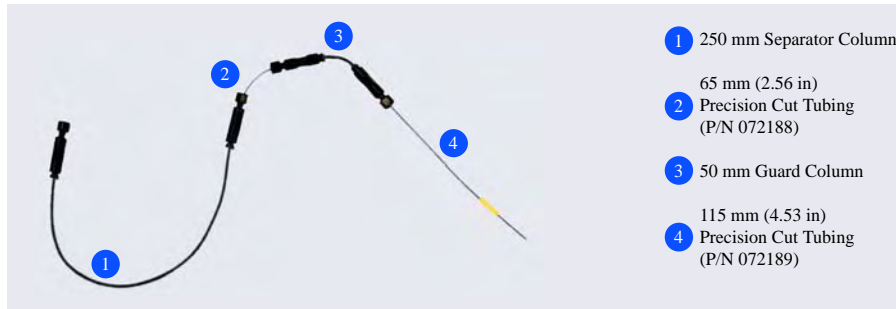


Figure 3
Tubing Connections for 250 mm Separator Column and 50 mm Guard Column



Figure 4
Tubing Connections for Separator Column Only

- Lift up the lid of the column cartridge to open it.
- Remove the fitting plug from the outlet fitting on the separator column. Orient the fitting with a flat side up (see Figure 5) and push the fitting into the opening at the front of the column cartridge until it stops.

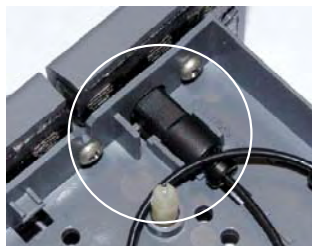


Figure 5
Column Outlet Fitting Installed in Column Cartridge

- Coil the separator column tubing inside the cartridge as shown in Figure 1 or Figure 2. Secure the column tubing and the inlet fitting in the clips on the column cartridge.
- Secure the inlet and outlet fittings on the guard column (if used) in the column clips on the lid of the column cartridge.
- Route the guard column inlet tubing (if used) or the separator column inlet tubing through the clip on the top edge of the column cartridge lid.
- Close the lid (you should hear a click) and route the tubing into the slot on the front of the column cartridge (see Figure 6).



NOTE

If the columns are installed correctly, the cartridge lid snaps closed easily. If the lid does not close easily, do not force it. Open the lid and verify that the columns and tubing are installed correctly and secured in the clips.



Figure 6
Column Cartridge Closed

SECTION 4 – OPERATION

4.1. General Operating Conditions

| | |
|---|---|
| Sample Volume: | 0.4-mm: 0.4 μ L Loop 2-mm: 2.5 μ L Loop + 0.8 μ L Injection valve dead volume 4-mm: 10 μ L Loop + 0.8 μ L Injection valve dead volume |
| Column: | 0.4-mm: Dionex IonPac AS11-HC 0.4-mm Capillary Column + Dionex IonPac AG11-HC 0.4-mm Guard Column 2-mm: Dionex IonPac AS11-HC 2-mm Analytical Column + Dionex IonPac AG11-HC 2-mm Guard Column 4-mm: Dionex IonPac AS11-HC 4-mm Analytical Column + Dionex IonPac AG11-HC 4-mm Guard Column |
| Eluent: | 30 mM NaOH (for test chromatogram) |
| Eluent Source: | Dionex EGC III KOH |
| Eluent Flow Rate: | 2-mm: 0.38 mL/min 4-mm: 1.5 mL/min 0.4-mm: 0.015 mL/min |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2- or 4-mm) Dionex Anion Capillary Electrolytic Suppression, ACES-300 AutoSuppression Recycle Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | < 3 μ S |
| Long-term Storage Solution (> 1 week): | 100 mM Sodium Borate |
| Short-term Storage Solution (< 1 week): | Eluent |

4.2. Dionex IonPac AS11-HC Operation Precautions

- a) Filter and Degas Eluents

Table 4
Filter and Degas Eluents

| FILTER SAMPLES | |
|--------------------------------------|-----------------------|
| Eluent pH | Between 0 and 14 |
| Sample pH | Between 0 and 14 |
| Maximum Flow Rate for 2 mm Columns | 0.75 mL/min |
| Maximum Flow Rate for 4 mm Columns | 3.0 mL/min |
| Maximum Flow Rate for 0.4 mm Columns | 0.03 mL/min |
| Maximum Operating Pressure | 4,000 psi (27.57 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. Thermo Fisher Scientific 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 Dionex IonPac AS11-HC 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 Thermo Fisher Scientific, we have obtained consistent results using 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 Dionex IonPac AS11-HC columns is 4,000 psi (27.57 MPa).

The Dionex IonPac AS11-HC 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 Dionex IonPac AS11-HC Columns

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

*Higher concentrations may only be used for limited duration applications such as column clean-up at pressures < 2000 psi.

**CAUTION**

The Dionex ASRS 300 and the Dionex ACES 300 must be operated in the AutoSuppression External Water Mode when using eluents containing solvents. Do not use > 40% solvent on the Dionex ASRS 300 and the Dionex ACES 300 in the electrolytic mode (power on).

4.4. Making Eluents that Contain Solvents

When mixing solvents with water remember to mix solvent with water on a volume to volume basis. For example, 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.

**NOTE**

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.

**NOTE**

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.

**SAFETY**

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

4.5. Eluent Preparation

4.5.1. Sodium Hydroxide Eluent Concentration

4.5.1.1. Weight Method

When formulating eluents from 50% sodium hydroxide, it is recommended to weigh out the required amount of 50% sodium hydroxide. Use the assayed concentration value from the sodium hydroxide bottle.

Example: To make 1 L of 30 mM NaOH use 2.4 g of 50% sodium hydroxide:
(as used in Section 5.3, “Production Test Chromatogram”)

$$\text{For 30 mM:} \quad \frac{0.03 \text{ mole/L} \times 40.01 \text{ g/mole}}{50\%} = 2.4 \text{ g diluted to 1 L}$$

4.5.1.2. Volume Method

Although it is more difficult to make precise carbonate-free eluents for gradient analysis volumetrically, you may choose to use the following formula to determine the correct volume of 50% sodium hydroxide to be diluted.

$$g = dvr$$

Where: **g** = weight of sodium hydroxide required (g)
d = density of the concentrated solution (g/mL)
v = volume of the 50% sodium hydroxide required (mL)
r = % purity of the concentrated solution

Example: To make 1 L of 20 mM NaOH use 1.05 mL of 50% sodium hydroxide:
(as used in Section 5.2, "Production Test Chromatogram")

For 20 mM: $\frac{0.03 \text{ mole/L} \times 40.01 \text{ g/mole}}{50\% \times 1.53^* \text{ g/mL}} = 1.57 \text{ mL diluted to 1 L}$

*This density applies to 50% NaOH. If the concentration of the NaOH solution is significantly different from 50%, the upper (weight method) calculation should be used instead.

4.5.1.3. Sodium Hydroxide Eluents

Dilute the amount of 50% (w/w) NaOH Reagent specified in Table 6, "Dilution of 50% (w/w) NaOH to Make Standard Dionex IonPac AS11-HC Eluents" with degassed, deionized water (18.2 megohm-cm) to a final volume of 1,000 mL using a volumetric flask. Avoid the introduction of carbon dioxide from the air into the aliquot of 50% (w/w) NaOH bottle or the deionized water being used to make the eluent. Do not shake the 50% (w/w) NaOH bottle or pipette the required aliquot from the top of the solution where sodium carbonate may have formed.

Table 6
Dilution of 50% (w/w) NaOH to Make
Standard Dionex IonPac AS11-HC Eluents

| 50% (w/w) NaOH g (mL) | | Concentration of NaOH Eluent (mM) |
|-----------------------------|----------|---|
| 0.08 | (0.0523) | 1 |
| 0.40 | (0.262) | 5 |
| 2.40 | (1.57) | 30 |
| 8.00 | (5.23) | 100 |
| 16.00 | (10.5) | 200 |
| 40.00 | (26.15) | 500 |

4.6. Regenerant Preparation for the Dionex AMMS 300

The Dionex Anion MicroMembrane Suppressor 300 (Dionex AMMS 300) requires the use of a regenerant solution. If you are using the Dionex AMMS 300 instead of the Dionex Anion Self-Regenerating Suppressor 300 (Dionex ASRS 300), the Product Manual for the Dionex AMMS 300 (Document No. 031727).

SECTION 5 – EXAMPLE APPLICATIONS

5.1. Recommendations for Optimum System Performance

The chromatograms in this section were obtained using columns that reproduced the Production Test Chromatogram on optimized Ion Chromatographs. 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.

The Dionex IonPac AS11-HC is designed to perform analyses of large numbers of anions of varying valencies through gradient elution. In any type of gradient elution system it is important to use eluents that produce a minimum shift in baseline conductivity during the run, as well as a fast equilibration time from one run to the next. Because sodium hydroxide is converted to water in the suppressor, it is the best choice for an eluent. As long as the capacity of the suppressor is not exceeded, the eluent hydroxide concentration has little effect on background conductivity. For example, a gradient run could begin at a few mM NaOH and end at 100 mM NaOH, with only a resulting 1 to 3 μ S total baseline change.

Ensure that your system is properly configured. It is very important that applications run on 2-mm columns utilize the proper pump configuration (see Section 2, “Ion Chromatography Systems”) and have all system void volumes minimized. Fluctuations in operating temperature can affect the retention time and resolution of analytes and should be controlled.

Ensure that adequate equilibration time is allowed between runs. If downward shift in baseline is observed during the isocratic section of the chromatogram, increase the equilibration time.

The addition of chromate to the sample will help stabilize organic acids. If your sample or standard contains organic acids, adding chromate (about 10 mg/L) will help stabilize them from bacterial degradation at room temperature. See the sample chromatograms in Sections 5.3, “Gradient Elution of a Large Number of Inorganic and Organic Acid Anions,” (Figure 9, “Separation of Mono-, Di-, and Trivalent Anions in One Sample Run”).

Use a guard column to protect the analytical/capillary column. If column performance deteriorates and it is determined that the guard and analytical/capillary columns has been fouled, refer to the column cleanup protocols in Appendix A, “Column Care.”

You can increase the sensitivity of your system by using sample concentration techniques (see Section 3.3, “The Sample Concentrator”).



CAUTION

Carbon dioxide readily dissolves in dilute basic solutions, forming carbonate. Carbonate contamination of eluents can affect the retention times of the anions being analyzed. Eluents should be maintained under an inert helium atmosphere to avoid carbonate contamination.

5.2. Isocratic Elution With and Without a Guard

Isocratic elution of common anions on the Dionex IonPac AS11-HC Analytical/Capillary Column has been optimized utilizing a hydroxide eluent. By using this eluent, common inorganic anions can be used to test the performance of the Dionex IonPac AS11-HC Column. The Dionex IonPac AS11-HC Analytical/Capillary Column should always be used with the Dionex IonPac AG11-HC Guard/Capillary Guard Column. An operating temperature of 30°C is used to ensure reproducible resolution and retention. Note that the Dionex IonPac AG11-HC Guard/Capillary Guard is packed with a microporous resin of proportionally lower capacity and contributes approximately 2% increase in retention times when a guard column is placed in-line prior to the analytical/capillary column.

Sample Volume: 2-mm: 2.5 μ L Loop + 0.8 μ L Injection valve dead volume
 4-mm: 10 μ L Loop + 0.8 μ L Injection valve dead volume

Column: See chromatogram

Eluent: 30 mM NaOH

Eluent Flow Rate: 0.38 mL/min (2-mm), 1.5 mL/min (4-mm)

Operating Temperature: 30°C

SRS Suppressor: Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm)
 AutoSuppression Recycle Mode

or MMS Suppressor: Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm)

MMS Regenerant: 50 mN H₂SO₄

Expected Background Conductivity: < 3 μ S

Long-term Storage Solution (> 1 week): 100 mM Sodium Borate

Short-term Storage Solution (< 1 week): Eluent

| Analyte | mg/L |
|--------------|------|
| 1. Fluoride | 2.0 |
| 2. Chloride | 5.0 |
| 3. Nitrite | 10.0 |
| 4. Sulfate | 10.0 |
| 5. Bromide | 20.0 |
| 6. Nitrate | 20.0 |
| 7. Phosphate | 30.0 |

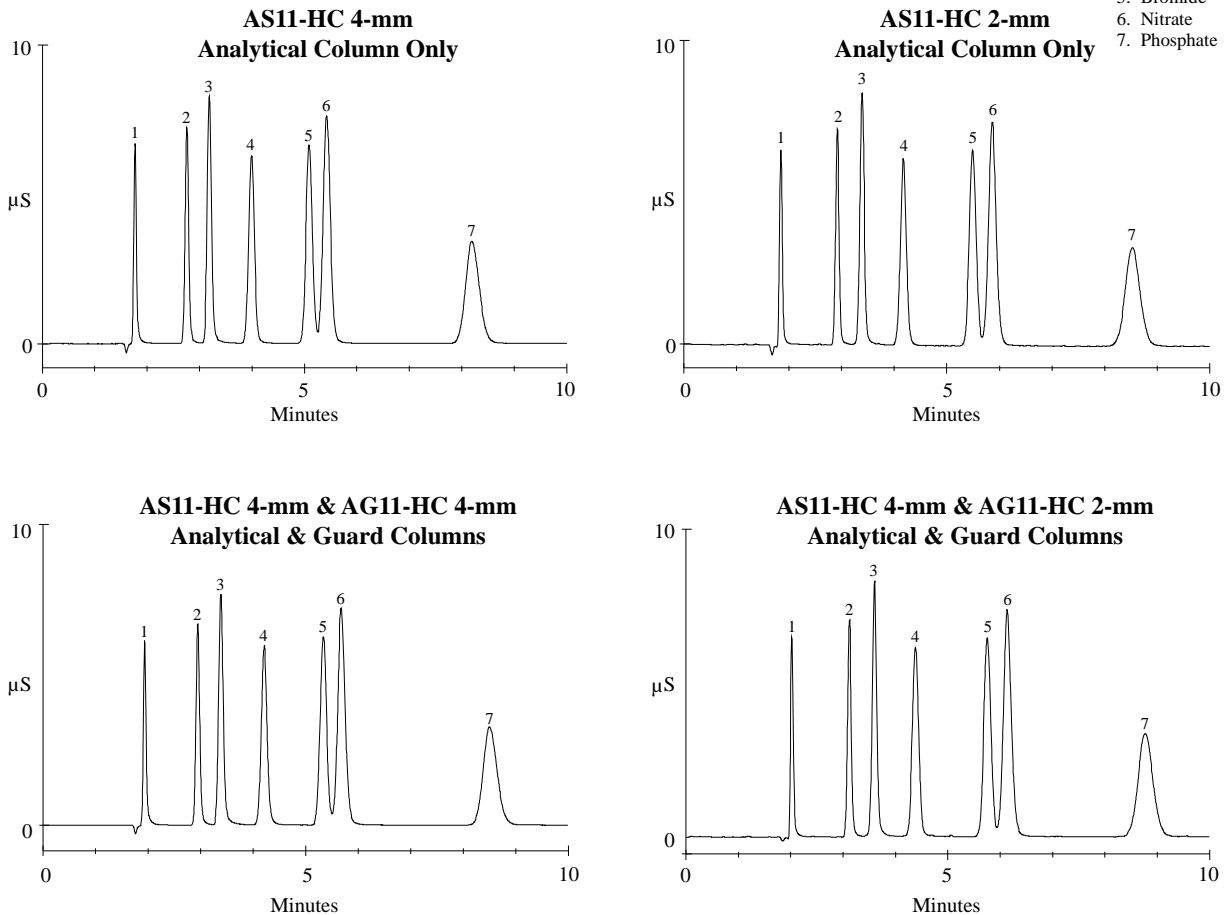


Figure 7A
Dionex IonPac AS11-HC Production Test Chromatograms

5.2.1. Isocratic Elution With and Without a Guard – Capillary

Column: Dionex IonPac AS11-HC (0.4 x 250 mm)
 Eluent Source: Dionex Capillary EGC-KOH Cartridge
 Eluent: Potassium hydroxide (EGC)
 30 mM
 Flow Rate: 0.015mL/min
 Inj. Volume: 0.4 µL
 Temperature: 30 C
 Detection: Suppressed conductivity, Dionex ACES 300, recycle mode

| Analyte | mg/L |
|--------------|------|
| 1. Fluoride | 0.5 |
| 2. Chloride | 1.25 |
| 3. Nitrite | 2.5 |
| 4. Sulfate | 2.5 |
| 5. Nitrate | 5 |
| 6. Bromide | 5 |
| 7. Phosphate | 7.5 |

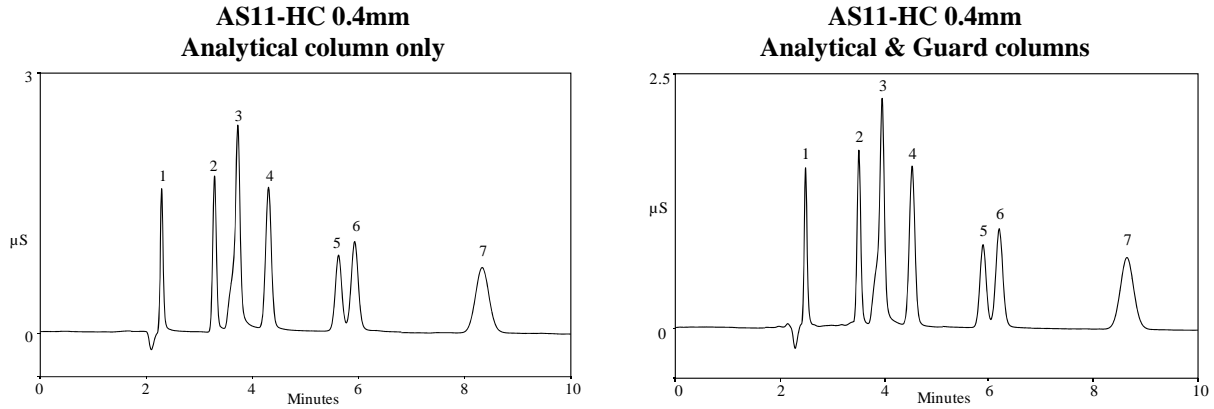


Figure 7B
Dionex IonPac AS11-HC Production Test Chromatogram – Capillary

5.3. Isocratic Elution at Room Temperature

Isocratic elution of seven common anions on the Dionex IonPac AS11-HC Analytical/Capillary Column has been optimized at 30°C. However, the column can be operated at room temperature. Notice that at room temperature (21°C) the divalent and trivalent ions have shorter retention times with 30 mM NaOH. The optimum eluent for room temperature operation is 25 mM NaOH.

| | |
|---|---|
| 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: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | See chromatogram |
| Eluent Flow Rate: | 0.38 mL/min (2-mm), 1.5 mL/min (4-mm) |
| Operating Temperature: | Room temperature (21°C) |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression Recycle Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | < 3 µS |
| Long-term Storage Solution (> 1 week): | 100 mM Sodium Borate |
| Short-term Storage Solution (< 1 week): | Eluent |

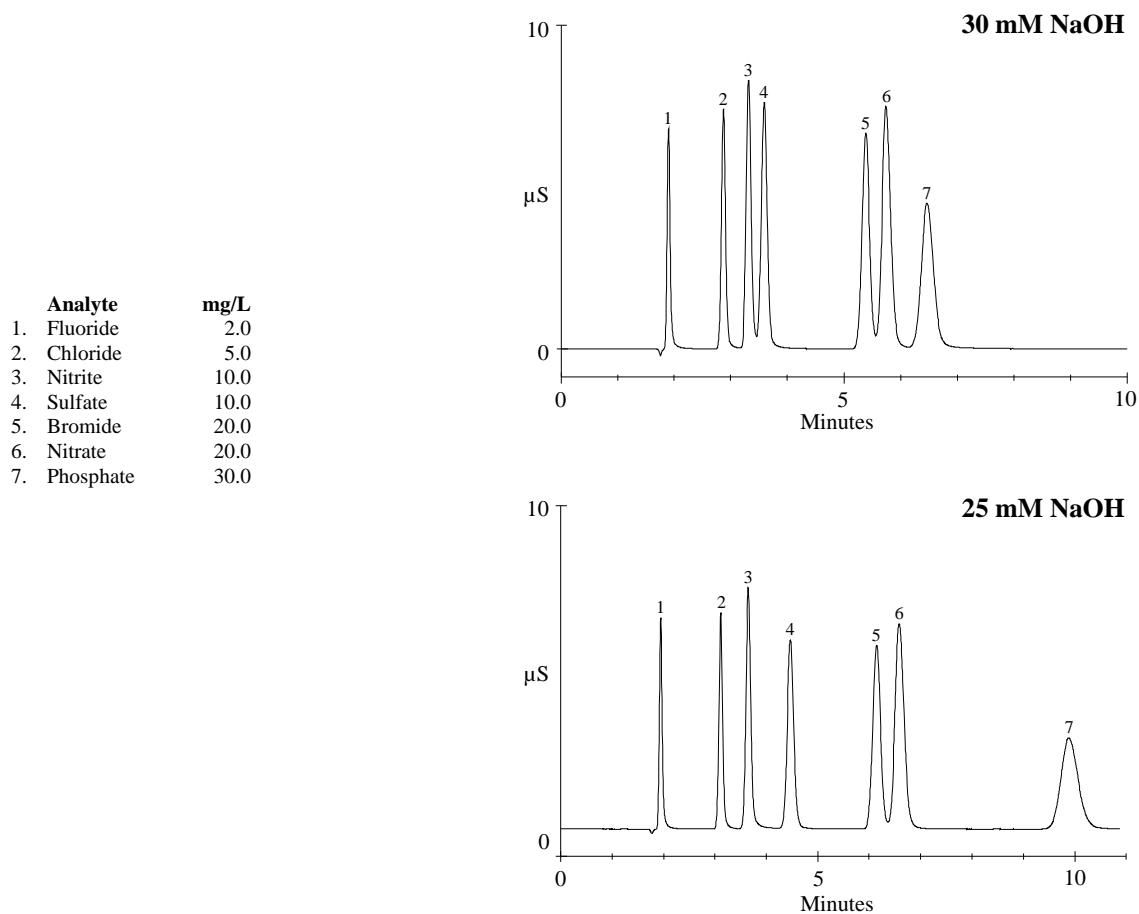


Figure 8
Dionex IonPac AS11-HC Test Chromatograms at Room Temperature

5.4. Gradient Elution of a Large Number of Inorganic Anions and Organic Acid Anions Using NaOH Gradient

A large number of inorganic anions and organic acid anions can be separated on the Dionex IonPac AS11-HC using gradient elution. The sodium hydroxide gradient is optimized in order to elute mono-, di-, and trivalent organic acid anions in a single run. The starting eluent in the beginning of the gradient has a low concentration allowing fluoride to elute after the void volume and also separates several weakly retained monovalent organic acids. The hydroxide concentration in the later part of the gradient elutes polyvalent ions such as trivalent phosphate, citrate, and cis- and trans-aconitate. See Section 5.2, "Eluent Preparation", for eluent preparation instructions.

Figure 9 demonstrates the effect of temperature on the selectivity of the Dionex IonPac AS11-HC. Operation at 30°C improves the separation of monovalents such as fluoride and lactate or formate and butyrate.

| | |
|-----------------------------------|---|
| Trap Column: | Dionex ATC-3, (Located between pump and injection valve) |
| Sample Volume: | 2-mm: 2.5 µL 4-mm: 10 µL |
| Column: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | E1: Deionized water E2: 5.0 mM NaOH E3: 100 mM NaOH E4: 100% Methanol |
| Eluent Flow Rate: | 0.38 mL/min (2-mm), 1.5 mL/min (4-mm) |
| Operating Temperature: | See Chromatogram |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression Recycle Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H2SO4 |
| Expected Background Conductivity: | 1.0 mM NaOH: 1 µS 60 mM NaOH: < 3.5 µS |
| Typical Operating Back Pressure: | 2200 psi (15.15 MPa) |

| TIME (min) | Gradient Conditions | | | | Comments |
|----------------------|---------------------|-----|-----|-----|-------------------------------|
| | %E1 | %E2 | %E3 | %E4 | |
| Equilibration | | | | | |
| 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 7 min |
| 7.0 | 80 | 20 | 0 | 0 | 7.0 |
| Analysis | | | | | |
| 7.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject |
| 7.5 | 80 | 20 | 0 | 0 | Inject Valve to Load Position |
| 15.0 | 80 | 20 | 0 | 0 | Isocratic analysis |
| 25.0 | 85 | 0 | 15 | 0 | Gradient analysis |
| 35.0 | 70 | 0 | 30 | 0 | |
| 45.0 | 40 | 0 | 60 | 0 | |
| 7.5 | 80 | 20 | 0 | 0 | Inject Valve to Load Position |
| 15.0 | 80 | 20 | 0 | 0 | Isocratic analysis |

| Analyte | mg/L | Analyte | mg/L | Analyte | mg/L |
|-----------------------|------|----------------------|------|---------------------|------|
| 1. Quinate | 10 | 18. Sorbate | 10 | 35. Phthalate | 20 |
| 2. Fluoride | 3 | 19. Trifluoroacetate | 10 | 36. Citrate | 20 |
| 3. Lactate | 10 | 20. Bromide | 10 | 37. Isocitrate | 20 |
| 4. Acetate | 10 | 21. Nitrate | 10 | 38. Chromate | 20 |
| 5. Glycolate | 10 | 22. Glutarate | 10 | 39. cis-Aconitate | 20 |
| 6. Propionate | 10 | 23. Carbonate | 20 | 40. trans-Aconitate | 20 |
| 7. Formate | 10 | 24. Succinate | 15 | | |
| 8. Butyrate | 10 | 25. Malate | 15 | | |
| 9. Methylsulfonate | 10 | 26. Malonate | 15 | | |
| 10. Pyruvate | 10 | 27. Tartrate | 15 | | |
| 11. Chlorite | 10 | 28. Maleate | 15 | | |
| 12. Valerate | 10 | 29. Sulfate | 15 | | |
| 13. Galacturonate | 10 | 30. Oxalate | 15 | | |
| 14. Monochloroacetate | 10 | 31. Fumarate | 15 | | |
| 15. Bromate | 10 | 32. Ketomalonate | 20 | | |
| 16. Chloride | 5 | 33. Tungstate | 20 | | |
| 17. Nitrite | 10 | 34. Phosphate | 20 | | |

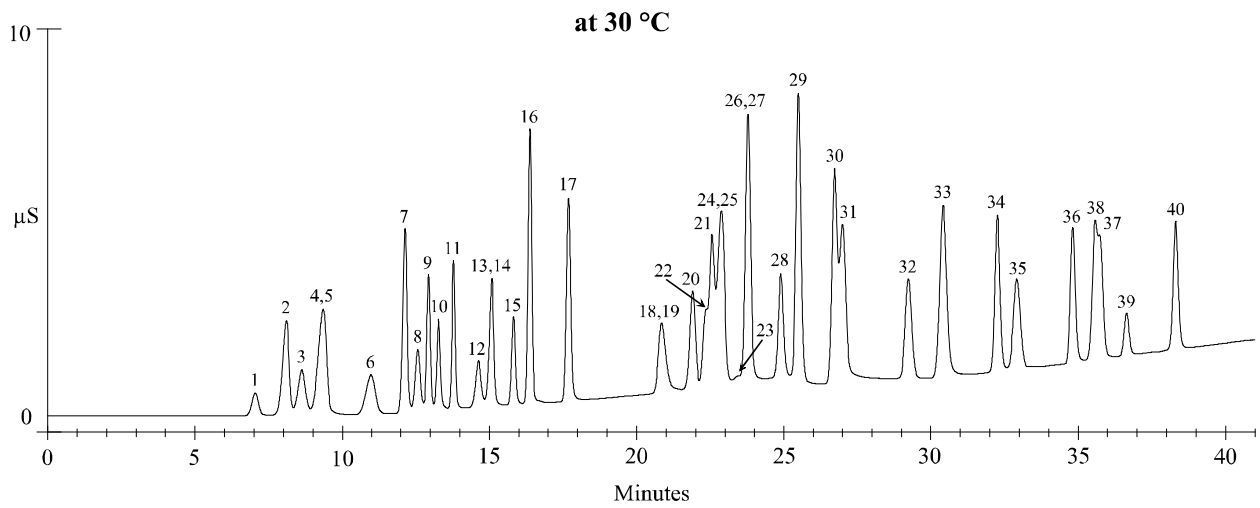
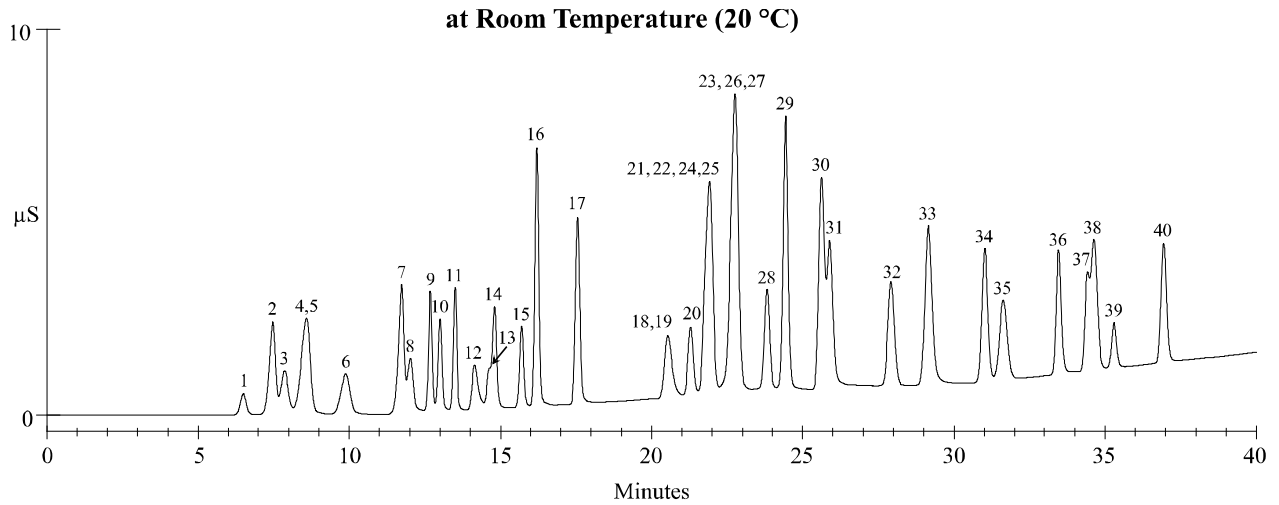


Figure 9
Gradient Elution of a Large Number of Inorganic Anions and
Organic Anions using NaOH Gradient

5.5. Gradient Elution of a Large Number of Inorganic Anions and Organic Acid Anions Using a NaOH Gradient without Solvent or with Solvent

Figure 10 illustrates the separation of a large number of inorganic anions and organic acids on the Dionex IonPac AS11-HC using a sodium hydroxide gradient. A solvent (methanol) gradient is used to optimize the resolution of closely eluting ions such as acetate and glycolate, succinate and malate, as well as malonate and tartrate. An operating temperature of 30°C is used to ensure reproducible resolution and retention times.

Trap Column: Dionex ATC-3, (Located between pump and injection valve)
 Sample Volume: 2-mm: 2.5 µL
 4-mm: 10 µL
 Column: Dionex IonPac AS11-HC Analytical and AG11-HC Guard
 Eluent: E1: Deionized water
 E2: 5.0 mM NaOH
 E3: 100 mM NaOH
 E4: 100% Methanol
 Eluent Flow Rate: 0.38 mL/min (2-mm), 1.5 mL/min (4-mm)
 Operating Temperature: 30°C
 SRS Suppressor: Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm)
 AutoSuppression External Water Mode
 or MMS Suppressor: Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm)
 MMS Regenerant: 50 mN H₂SO₄
 Expected Background Conductivity: 1.0 mM NaOH: 1 µS 60 mM NaOH: < 3.5 µS
 Typical Operating Back Pressure: Without Solvent: 2,200 psi (15.15 MPa)
 With Solvent: 2,700 psi (18.60 MPa)

| Gradient Conditions Without Solvent | | | | | | Gradient Conditions With Solvent | | | | | |
|-------------------------------------|-----|-----|-----|-----|-------------------------------|----------------------------------|-----|-----|-----|-----|-------------------------------|
| TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments | TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments |
| Equilibration | | | | | | Equilibration | | | | | |
| 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 7 min | 0 | 70 | 20 | 0 | 10 | 1.0 mM NaOH for 7 min |
| 7.0 | 80 | 20 | 0 | 0 | 7.0 | 7.0 | 70 | 20 | 0 | 10 | 7.0 |
| Analysis | | | | | | Analysis | | | | | |
| 7.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject | 7.1 | 70 | 20 | 0 | 10 | 1.0 mM NaOH, Inject |
| 7.5 | 80 | 20 | 0 | 0 | Inject Valve to Load Position | 7.5 | 70 | 20 | 0 | 10 | Inject Valve to Load Position |
| 15.0 | 80 | 20 | 0 | 0 | Isocratic analysis | 15.0 | 70 | 20 | 0 | 10 | Isocratic analysis |
| 25.0 | 85 | 0 | 15 | 0 | Gradient analysis | 25.0 | 65 | 0 | 15 | 20 | Gradient analysis |
| 35.0 | 70 | 0 | 30 | 0 | | 35.0 | 50 | 0 | 30 | 20 | |
| 45.0 | 40 | 0 | 60 | 0 | | 45.0 | 30 | 0 | 60 | 10 | |

| Analyte | mg/L | Analyte | mg/L | Analyte | mg/L |
|-----------------------|------|----------------------|------|---------------------|------|
| 1. Quinate | 10 | 15. Bromate | 10 | 29. Sulfate | 15 |
| 2. Fluoride | 3 | 16. Chloride | 5 | 30. Oxalate | 15 |
| 3. Lactate | 10 | 17. Nitrite | 10 | 31. Fumarate | 15 |
| 4. Acetate | 10 | 18. Sorbate | 10 | 32. Ketomalonate | 20 |
| 5. Glycolate | 10 | 19. Trifluoroacetate | 10 | 33. Tungstate | 20 |
| 6. Propionate | 10 | 20. Bromide | 10 | 34. Phosphate | 20 |
| 7. Formate | 10 | 21. Nitrate | 10 | 35. Phthalate | 20 |
| 8. Butyrate | 10 | 22. Glutarate | 10 | 36. Citrate | 20 |
| 9. Methylsulfonate | 10 | 23. Carbonate | 20 | 37. Isocitrate | 20 |
| 10. Pyruvate | 10 | 24. Succinate | 15 | 38. Chromate | 20 |
| 11. Chlorite | 10 | 25. Malate | 15 | 39. cis-Aconitate | 20 |
| 12. Valerate | 10 | 26. Malonate | 15 | 40. trans-Aconitate | |
| 13. Galacturonate | 10 | 27. Tartrate | 15 | | |
| 14. Monochloroacetate | 10 | 28. Maleate | 15 | | |

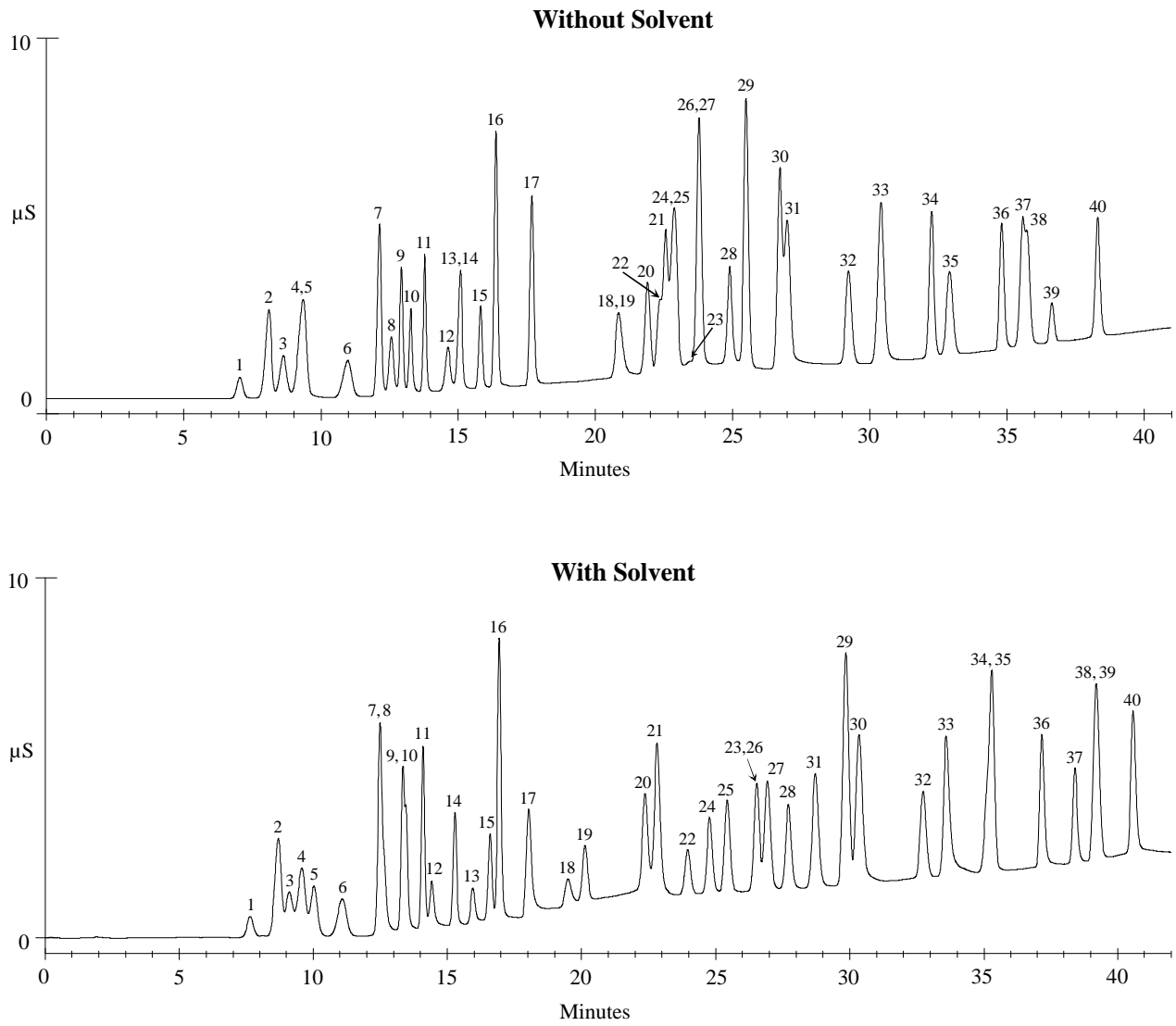


Figure 10
Gradient Analysis of a Large Number of Anions
using NaOH without Solvent and with Solvent

5.6. Comparison of Dionex IonPac AS11-HC 4 mm and 0.4 mm Columns

Column: Dionex IonPac AG11-HC/AS11-HC (see chromatogram)
 Eluent Source: Dionex EGC-KOH Cartridge
 Eluent (4 mm): Potassium hydroxide:
 1mM from 0 to 7 min, 1mM to 15mM from 7 to 16 min, 15mM to 30mM from 16 to 25 min, 30mM to 60mM from 25 to 33 min
 Eluent (0.4 mm): Potassium hydroxide:
 1mM from 0 to 5 min, 1mM to 15mM from 5 to 14 min, 15mM to 30mM from 14 to 23 min, 30mM to 60mM from 23 to 31 min
 Flow Rate: 1.5 mL/min (4 mm), 0.015 mL/min (0.4 mm)
 Inj. Volume: 40 µL (4 mm), 0.40 µL (0.4 mm)
 Temperature: 30 °C
 Detection: Suppressed conductivity, Dionex ASRS 300 4mm Dionex ACES 300 0.4mm recycle mode

Note: The isocratic time was reduced by two minutes for capillary AS11-HC 0.4mm method due to the difference in system delay volume for analytical and capillary systems.

| Peak | mg/L | Peak | mg/L |
|-----------------------|------|---------------------|------|
| 1. Quinate | 5.0 | 16. Bromide | 5.0 |
| 2. Fluoride | 1.5 | 17. Nitrate | 5.0 |
| 3. Lactate | 5.0 | 18. Carbonate | --- |
| 4. Acetate | 5.0 | 19. Malonate | 7.5 |
| 5. Propionate | 5.0 | 20. Maleate | 7.5 |
| 6. Formate | 5.0 | 21. Sulfate | 7.5 |
| 7. Butyrate | 5.0 | 22. Oxalate | 7.5 |
| 8. Methylsulfonate | 5.0 | 23. Tungstate | 10.0 |
| 9. Pyruvate | 5.0 | 24. Phosphate | 10.0 |
| 10. Valerate | 5.0 | 25. Pthalate | 10.0 |
| 11. Monochloroacetate | 5.0 | 26. Citrate | 10.0 |
| 12. Bromate | 2.5 | 27. Chromate | 10.0 |
| 14. Nitrite | 5.0 | 28. cis-Aconitate | --- |
| 13. Chloride | 5.0 | 29. trans-Aconitate | 10.0 |
| 15. Trifluoroacetate | 5.0 | | |

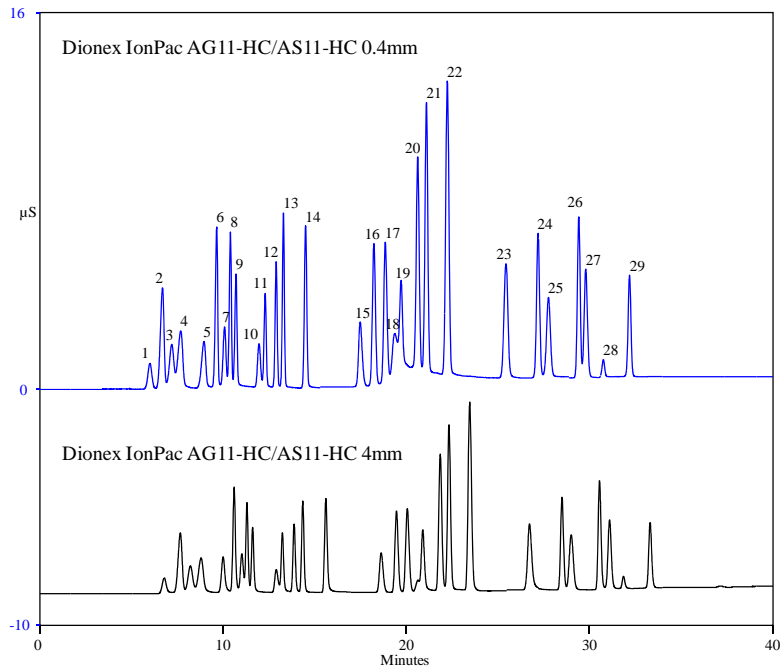


Figure 11
Comparison of Dionex IonPac AS11-HC 4 mm and 0.4 mm Columns

5.7. Method Optimization for Food and Beverage Applications Using NaOH gradient with a Solvent Step Change or a Solvent Gradient

Figure 12 illustrates the use of a solvent gradient compared to a step change to optimize peak resolution. These examples illustrate how a solvent gradient or step change can be introduced at different points in the gradient to modify column selectivity for the analytes. Note that in examples A and B, good resolution of lactate/acetate and formate/butyrate can be achieved in the beginning section of the chromatogram without solvent. Resolution of succinate and malate is optimized by the addition of solvent with a linear solvent gradient shown in example B. Sulfate and oxalate resolution is optimized by using a solvent step change, shown in example A.

| | |
|-----------------------------------|--|
| Trap Column: | Dionex ATC-3, (Located between pump and injection valve) |
| Sample Volume: | 2-mm: 2.5 µL 4-mm: 10 µL |
| Column: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | E1: Deionized water E2: 5.0 mM NaOH E3: 100 mM NaOH E4: 100% Methanol |
| Eluent Flow Rate: | 0.38 mL/min (2-mm), 1.5 mL/min (4-mm) |
| Operating Temperature: | 30°C |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression External Water Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | 1.0 mM NaOH: 1 µS 60 mM NaOH: < 3.5 µS |
| Typical Operating Back Pressure: | Without Solvent: 2,200 psi (15.15 MPa) With Solvent: 2,700 psi (18.60 MPa) |

| Example A Gradient Conditions | | | | | | Example B Gradient Conditions | | | | | |
|----------------------------------|-----|-----|-----|-----|-------------------------------|----------------------------------|-----|-----|-----|-----|---------------------------------|
| TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments | TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments |
| Equilibration | | | | | | Equilibration | | | | | |
| 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 8 min | 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 8 min |
| 8.0 | 80 | 20 | 0 | 0 | | 8.0 | 80 | 20 | 0 | 0 | |
| Analysis | | | | | | Analysis | | | | | |
| 8.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject | 8.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject |
| 8.5 | 80 | 20 | 0 | 0 | Inject Valve to Load Position | 8.5 | 80 | 20 | 0 | 0 | Inject Valve to Load Position |
| 16.0 | 80 | 20 | 0 | 0 | Isocratic analysis | 16.0 | 80 | 20 | 0 | 0 | End isocratic analysis |
| 22.0 | 90 | 0 | 10 | 0 | NaOH gradient | 26.0 | 65 | 0 | 15 | 20 | Methanol /NaOH gradient |
| 22.1 | 72 | 0 | 10 | 18 | Methanol step change | 36.0 | 50 | 0 | 30 | 20 | Start reverse methanol gradient |
| 26.0 | 67 | 0 | 15 | 18 | | 46.0 | 40 | 0 | 60 | 0 | |
| 36.0 | 22 | 0 | 60 | 18 | | | | | | | |
| 46.0 | 22 | 0 | 60 | 18 | | | | | | | |

| Analyte | mg/L | Analyte | mg/L | Analyte | mg/L |
|-----------------------|------|----------------------|------|---------------------|------|
| 1. Quinate | 10 | 15. Bromate | 10 | 29. Sulfate | 15 |
| 2. Fluoride | 3 | 16. Chloride | 5 | 30. Oxalate | 15 |
| 3. Lactate | 10 | 17. Nitrite | 10 | 31. Fumarate | 15 |
| 4. Acetate | 10 | 18. Sorbate | 10 | 32. Ketomalonate | 20 |
| 5. Glycolate | 10 | 19. Trifluoroacetate | 10 | 33. Tungstate | 20 |
| 6. Propionate | 10 | 20. Bromide | 10 | 34. Phosphate | 20 |
| 7. Formate | 10 | 21. Nitrate | 10 | 35. Phthalate | 20 |
| 8. Butyrate | 10 | 22. Glutarate | 10 | 36. Citrate | 20 |
| 9. Methylsulfonate | 10 | 23. Carbonate | 20 | 37. Isocitrate | 20 |
| 10. Pyruvate | 10 | 24. Succinate | 15 | 38. Chromate | 20 |
| 11. Chlorite | 10 | 25. Malate | 15 | 39. cis-Aconitate | 20 |
| 12. Valerate | 10 | 26. Malonate | 15 | 40. trans-Aconitate | |
| 13. Galacturonate | 10 | 27. Tartrate | 15 | | |
| 14. Monochloroacetate | 10 | 28. Maleate | 15 | | |

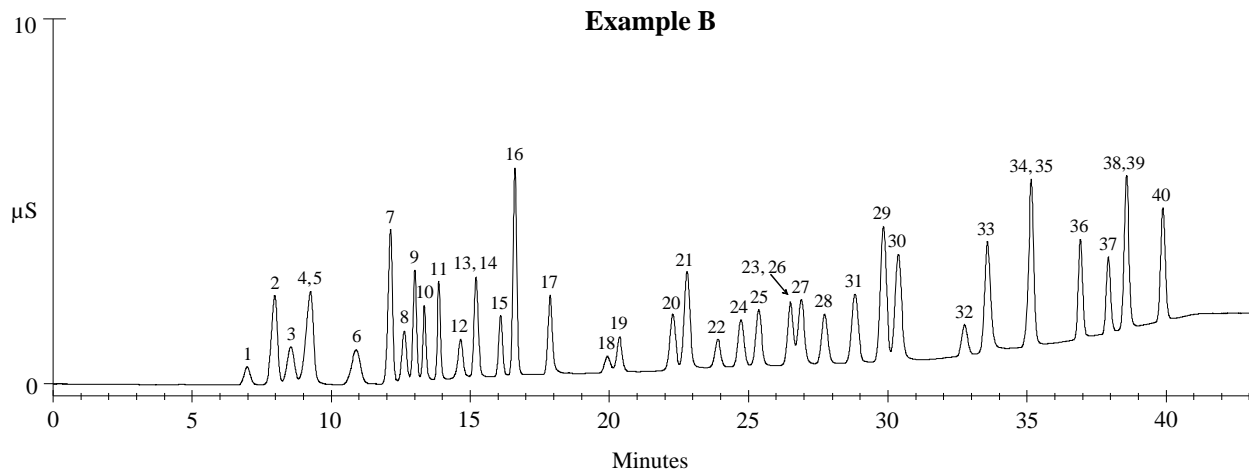
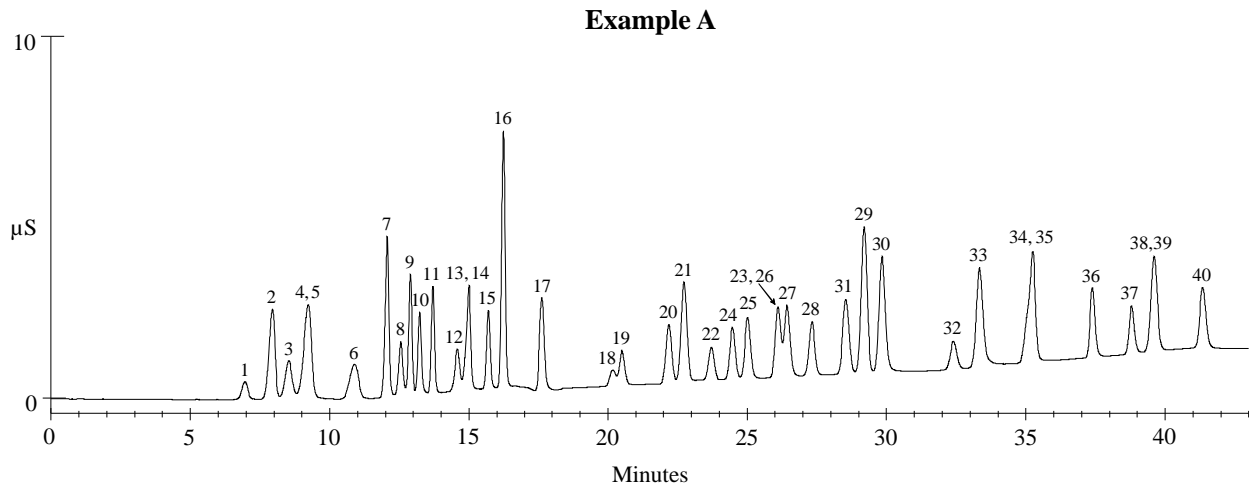


Figure 12
Elution of a Large Number of Inorganic Anions and Organic Acid Anions
Using NaOH Gradient with Solvent Step Change or Linear Solvent Gradient

5.8. Analysis of a Beer Sample

Figure 13 uses an optimized sodium hydroxide gradient with a methanol step gradient for analysis of a beer sample. The beer sample was diluted 1:5 with deionized water and treated with a Thermo Scientific Dionex OnGuard RP cartridge. The Dionex IonPac AS11-HC column has a high capacity which allows injection of concentrated samples for determination of trace components.

Dionex OnGuard RP Sample Pretreatment Procedure

1. Wash Dionex OnGuard RP with 10 mL methanol
2. Next wash with 10 mL deionized water
3. Discard 3-4 mL diluted sample then collect next 6-7 mL sample.

| | |
|-----------------------------------|--|
| Trap Column: | Dionex ATC-3, (Located between pump and injection valve) |
| Sample Volume: | 2-mm: 2.5 μ L 4-mm: 10 μ L |
| Column: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | E1: Deionized water E2: 5.0 mM NaOH E3: 100 mM NaOH E4: 100% Methanol |
| Eluent Flow Rate: | 0.38 mL/min (2-mm), 1.5 mL/min (4-mm) |
| Operating Temperature: | 30°C |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression External Water Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | 1.0 mM NaOH: 1 μ S 60 mM NaOH: < 3.5 μ S |
| Typical Operating Back Pressure: | 2700 psi (18.60 MPa) |

| Gradient Conditions | | | | | | Analyte |
|----------------------|-----|-----|-----|-----|---|---------------------|
| TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments | |
| Equilibration | | | | | | 1. Lactate |
| 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 8 min | 2. Acetate |
| 8.0 | 80 | 20 | 0 | 0 | | 3. Formate |
| Analysis | | | | | | 4. Butyrate |
| 8.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject Inject Valve to Load Position | 5. Pyruvate |
| 8.5 | 80 | 20 | 0 | 0 | | 6. Chloride |
| 16.0 | 80 | 20 | 0 | 0 | End isocratic analysis | 7. Bromide |
| 22.0 | 90 | 0 | 10 | 0 | NaOH gradient | 8. Nitrate |
| 22.1 | 72 | 0 | 10 | 18 | Methanol step change | 9. Succinate |
| 26.0 | 67 | 0 | 15 | 18 | | 10. Malate |
| 36.0 | 22 | 0 | 60 | 18 | | 11. Carbonate |
| 46.0 | 22 | 0 | 60 | 18 | | 12. Fumarate |
| | | | | | | 13. Sulfate |
| | | | | | | 14. Oxalate |
| | | | | | | 15. Phosphate |
| | | | | | | 16. Citrate |
| | | | | | | 17. Isocitrate |
| | | | | | | 18. cis-Aconitate |
| | | | | | | 19. trans-Aconitate |

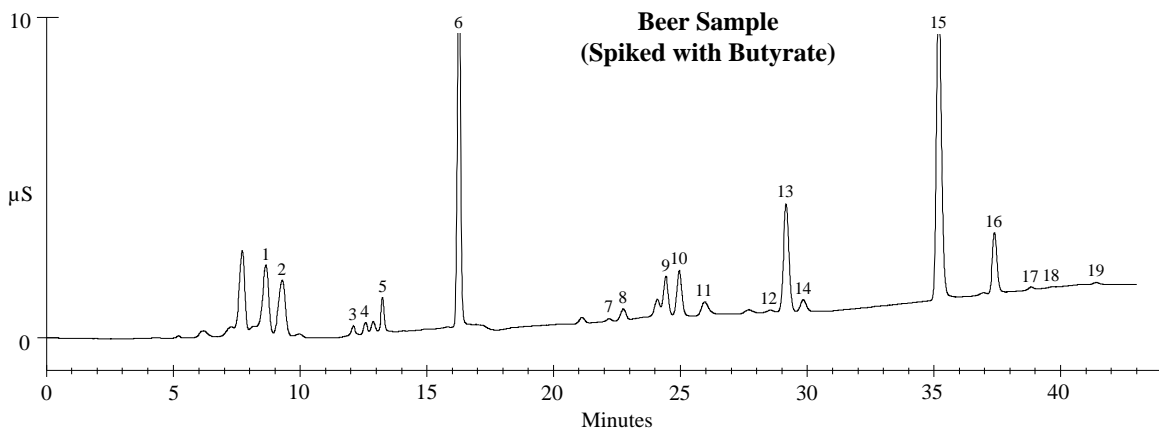
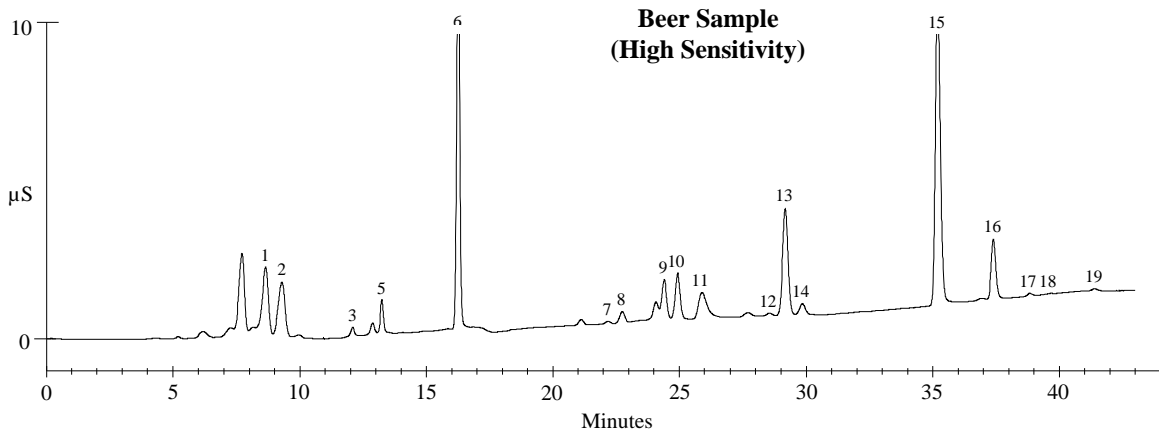
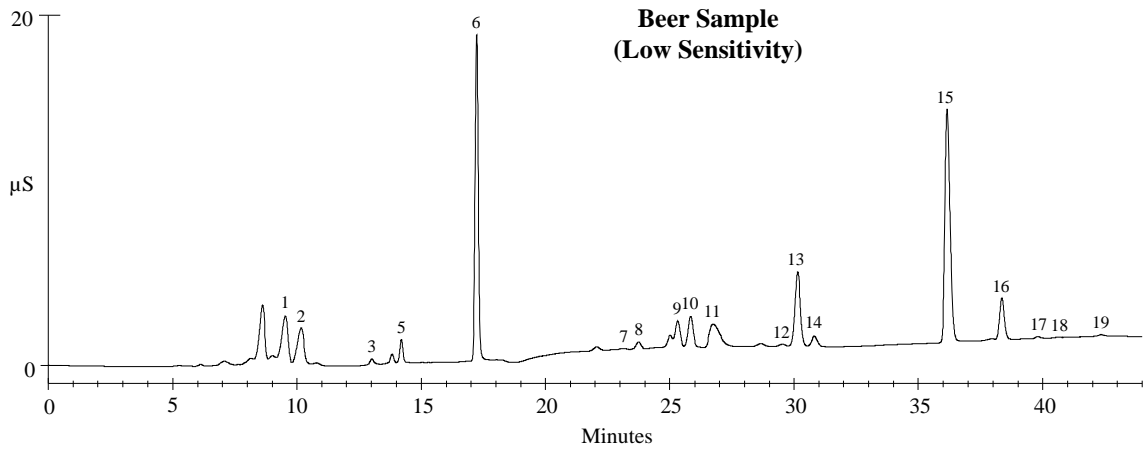


Figure 13
Analysis of a Beer Sample

5.9. Analysis of Apple Juice Samples

Figure 14 uses an optimized sodium hydroxide gradient with a methanol step gradient for analysis of apple juice samples. All three juice samples were diluted 1:10 with deionized water and filtered through a 0.45 mm syringe filter.

| | |
|-----------------------------------|--|
| Trap Column: | Dionex ATC-3, (Located between pump and injection valve) |
| Sample Volume: | 2-mm: 2.5 μ L 4-mm: 10 μ L |
| Column: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | E1: Deionized water E2: 5.0 mM NaOH E3: 100 mM NaOH E4: 100% Methanol |
| Eluent Flow Rate: | 0.38 mL/min (2-mm), 1.5 mL/min (4-mm) |
| Operating Temperature: | 30°C |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression External Water Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | 1.0 mM NaOH: 1 μ S 60 mM NaOH: < 3.5 μ S |
| Typical Operating Back Pressure: | 2700 psi (18.60 MPa) |

| Gradient Conditions | | | | | | Analyte |
|----------------------|-----|-----|-----|-----|-------------------------------|-------------------|
| TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments | |
| Equilibration | | | | | | 1. Quinate |
| 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 8 min | 2. Lactate |
| 8.0 | 80 | 20 | 0 | 0 | | 3. Acetate |
| Analysis | | | | | | 4. Propionate |
| 8.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject | 5. Formate |
| 8.5 | 80 | 20 | 0 | 0 | Inject Valve to Load Position | 6. Galacturonate |
| 16.0 | 80 | 20 | 0 | 0 | End isocratic analysis | 7. Chloride |
| 22.0 | 90 | 0 | 10 | 0 | NaOH gradient | 8. Nitrate |
| 22.1 | 72 | 0 | 10 | 18 | Methanol step change | 9. Succinate |
| 26.0 | 67 | 0 | 15 | 18 | | 10. Malate |
| 36.0 | 22 | 0 | 60 | 18 | | 11. Fumarate |
| 46.0 | 22 | 0 | 60 | 18 | | 12. Sulfate |
| | | | | | | 13. Oxalate |
| | | | | | | 14. Phosphate |
| | | | | | | 15. Citrate |
| | | | | | | 16. Isocitrate |
| | | | | | | 17. cis-Aconitate |

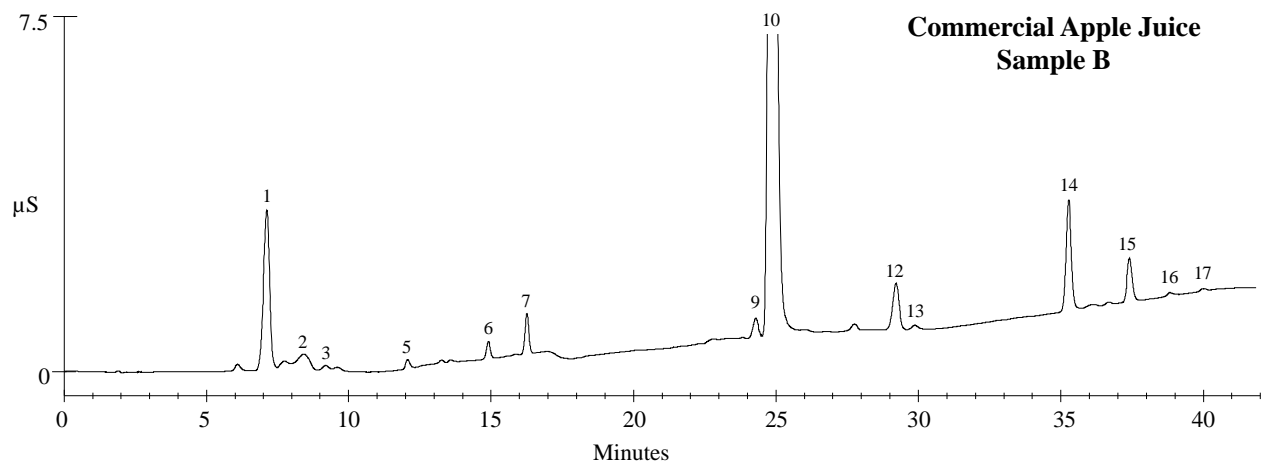
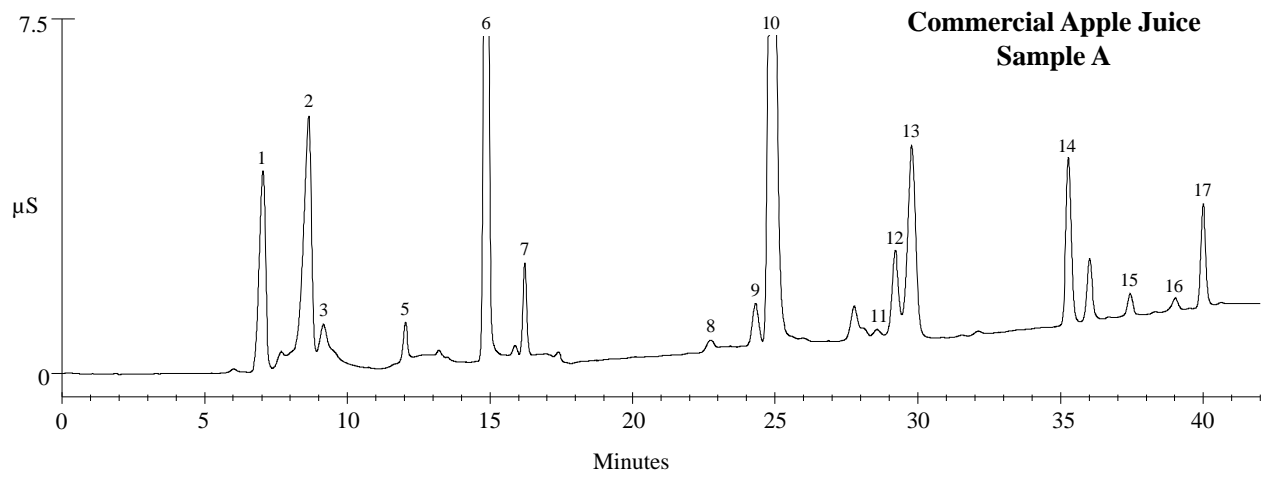
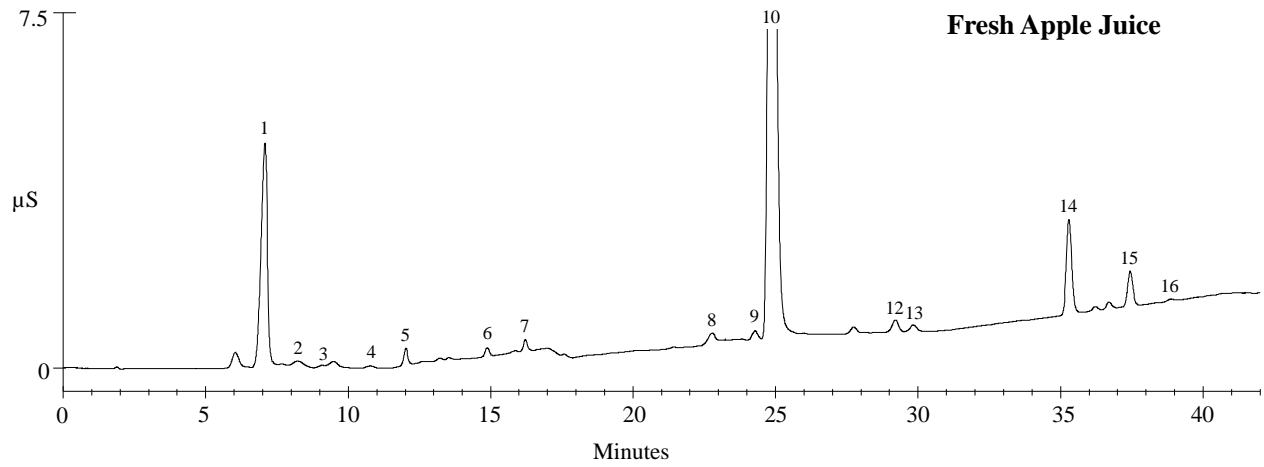


Figure 14
Analysis of Apple Juice Samples

5.10. Analysis of Orange Juice Samples

Figure 15 uses an optimized sodium hydroxide gradient with a methanol step gradient for analysis of an orange juice sample. Both juice samples were diluted 1:10 with deionized water and filtered through a 0.45 mm syringe filter.

| | |
|-----------------------------------|--|
| Trap Column: | Dionex ATC-3, (Located between pump and injection valve) |
| Sample Volume: | 2.5 μ L (2-mm), 10 μ L (4-mm) |
| Column: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | E1: Deionized water E2: 5.0 mM NaOH E3: 100 mM NaOH E4: 100% Methanol |
| Eluent Flow Rate: | 0.38 mL/min (2-mm), 1.5 mL/min (4-mm) |
| Operating Temperature: | 30°C |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression External Water Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | 1.0 mM NaOH: 1 μ S 60 mM NaOH: < 3.5 μ S |
| Typical Operating Back Pressure: | 2700 psi (18.60 MPa) |

| TIME (min) | Gradient Conditions | | | | Comments | Analyte |
|----------------------|---------------------|-----|-----|-----|-------------------------------|---------------------|
| | %E1 | %E2 | %E3 | %E4 | | |
| Equilibration | | | | | | |
| 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 8 min | 1. Quinate |
| 8.0 | 80 | 20 | 0 | 0 | | 2. Lactate |
| Analysis | | | | | | 3. Acetate |
| 8.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject | 4. Propionate |
| 8.5 | 80 | 20 | 0 | 0 | Inject Valve to Load Position | 5. Formate |
| 16.0 | 80 | 20 | 0 | 0 | Isocratic analysis | 6. Galacturonate |
| 22.0 | 90 | 0 | 10 | 0 | NaOH gradient | 7. Chloride |
| 22.1 | 72 | 0 | 10 | 18 | Methanol step change | 8. Glutarate |
| 26.0 | 67 | 0 | 15 | 18 | | 9. Succinate |
| 36.0 | 22 | 0 | 60 | 18 | | 10. Malate |
| 46.0 | 22 | 0 | 60 | 18 | | 11. Malonate |
| | | | | | | 12. Sulfate |
| | | | | | | 13. Oxalate |
| | | | | | | 14. Phosphate |
| | | | | | | 15. Citrate |
| | | | | | | 16. Isocitrate |
| | | | | | | 17. Trans-Aconitate |

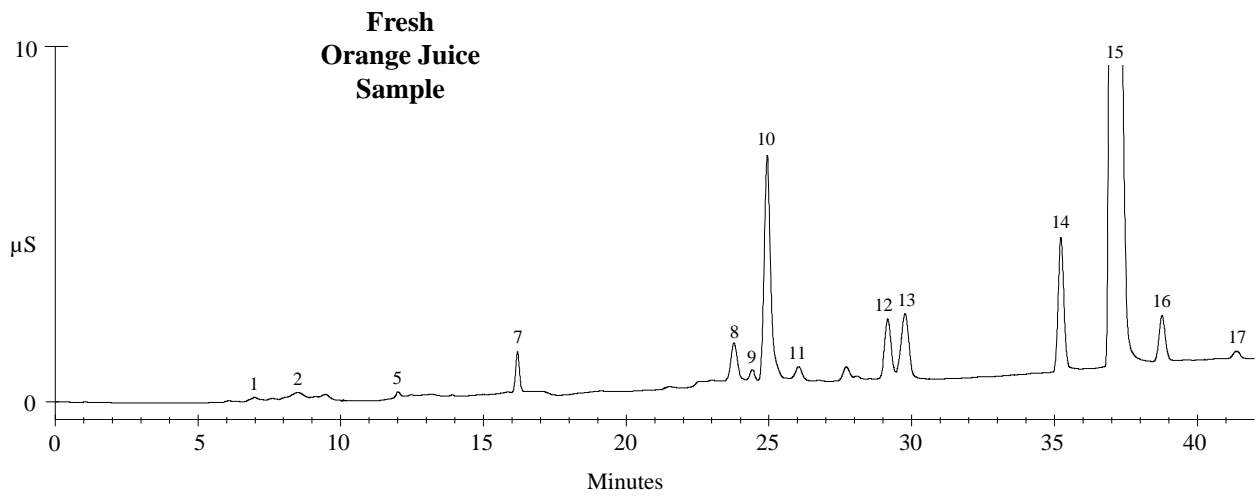
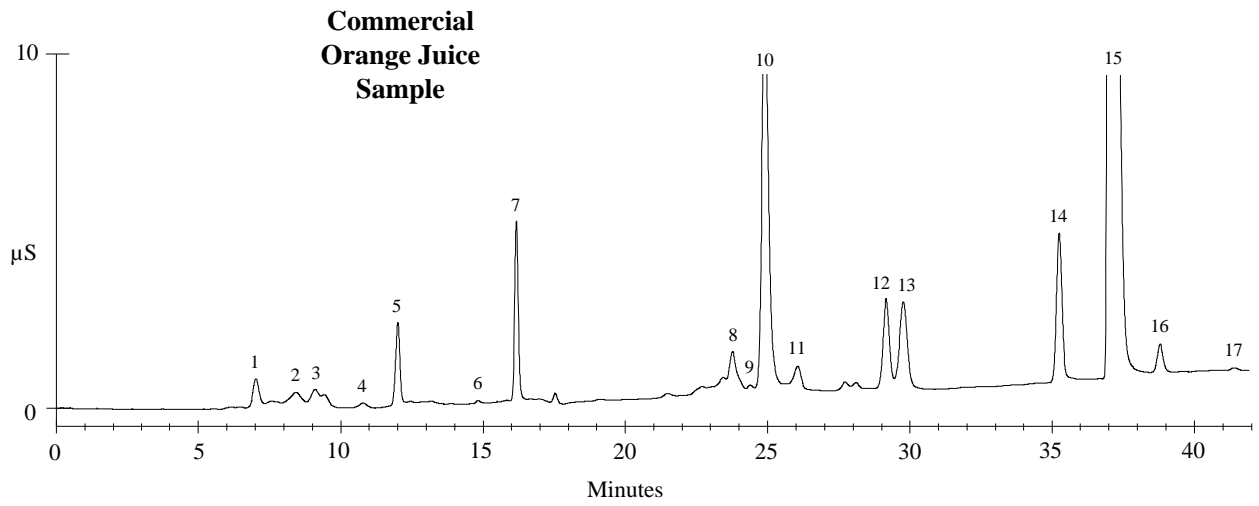


Figure 15
Analysis of Orange Juice Samples

5.11. Analysis of a Wine Sample

Figure 16 uses an optimized sodium hydroxide gradient with a linear methanol gradient for the analysis of red wine. The wine samples were diluted with deionized water and treated with a Dionex OnGuard RP cartridge.

Dionex OnGuard RP Sample Pretreatment Procedure

1. Wash Dionex OnGuard RP with 10 mL methanol
2. Next wash with 10 mL deionized water
3. Discard 3-4 mL diluted sample then collect next 6-7 mL sample.

| | |
|-----------------------------------|--|
| Trap Column: | Dionex ATC-3, (Located between pump and injection valve) |
| Sample Volume: | 2.5 μ L (2-mm), 10 μ L (4-mm) |
| Sample Dilution: | Sample A: 1:20 Sample B: 1:10 |
| Column: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | E1: Deionized water E2: 5.0 mM NaOH E3: 100 mM NaOH E4: 100% Methanol |
| Eluent Flow Rate: | 0.38 mL/min (2-mm), 1.5 mL/min (4-mm) |
| Operating Temperature: | 30°C |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression External Water Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | 1.0 mM NaOH: 1 μ S 60 mM NaOH: < μ S |
| Typical Operating Back Pressure: | 2700 psi (18.60 MPa) |

Gradient Conditions

| TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments |
|----------------------|-----|-----|-----|-----|-------------------------------|
| Equilibration | | | | | |
| 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 8 min |
| 8.0 | 80 | 20 | 0 | 0 | |
| Analysis | | | | | |
| 8.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject |
| 8.5 | 80 | 20 | 0 | 0 | Inject Valve to Load Position |
| 16.0 | 80 | 20 | 0 | 0 | Isocratic analysis |
| 26.0 | 65 | 0 | 15 | 20 | Methanol/NaOH gradient |
| 36.0 | 50 | 0 | 30 | 20 | |
| 46.0 | 40 | 0 | 60 | 0 | |

Analyte

1. Lactate
2. Acetate
3. Formate
4. Pyruvate
5. Galacturonate
6. Chloride
7. Bromide
8. Nitrate
9. Succinate
10. Malate
11. Tartrate
12. Sulfate
13. Oxalate
14. Phosphate
15. Citrate
16. Isocitrate
17. cis-Aconitate

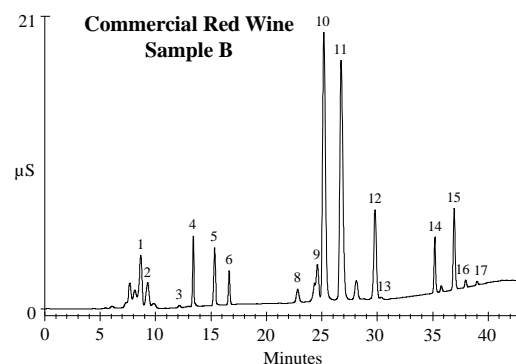
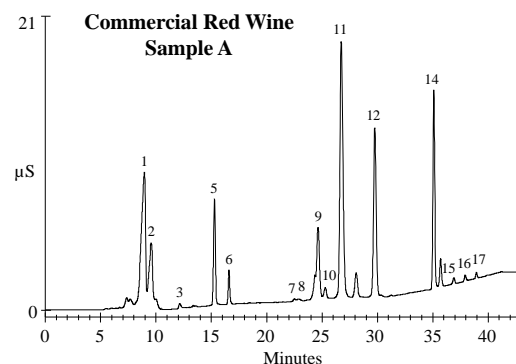


Figure 16
Isocratic Analysis of a Wine Sample

5.12. Gradient Elution of Organic Acid Anions, Common Inorganic Anions, and Hydrophobic Inorganic Anions

Figure 17 illustrates an optimized separation of highly retained hydrophobic inorganic anions. This method allows the resolution of acetate, glycolate, propionate, and formate while eluting thiosulfate, iodide, and thiocyanate in less than 45 minutes.

| | |
|-----------------------------------|--|
| Trap Column: | Dionex ATC-3, (Located between pump and injection valve) |
| Sample Volume: | 2.5 μ L (2-mm), 10 μ L (4-mm) |
| Column: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | E1: Deionized water E2: 5.0 mM NaOH E3: 100 mM NaOH E4: 100% Methanol |
| Eluent Flow Rate: | 0.38 mL/min (2-mm), 1.5 mL/min (4-mm) |
| Operating Temperature: | 30°C |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression External Water Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | 1.0 mM NaOH: 1 μ S 50 mM NaOH: < 3.5 μ S |
| Typical Operating Back Pressure: | 2700 psi (18.60 MPa) |

Gradient Conditions

| TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments |
|----------------------|-----|-----|-----|-----|-------------------------------|
| Equilibration | | | | | |
| 0 | 65 | 20 | 0 | 15 | 1.0 mM NaOH for 7 min |
| 8.0 | 65 | 20 | 0 | 15 | |
| Analysis | | | | | |
| 7.1 | 65 | 20 | 0 | 15 | 1.0 mM NaOH, Inject |
| 7.5 | 65 | 20 | 0 | 15 | Inject Valve to Load Position |
| 16.0 | 65 | 20 | 0 | 15 | End isocratic analysis |
| 24.0 | 70 | 0 | 15 | 15 | Gradient analysis |
| 32.0 | 55 | 0 | 30 | 15 | |
| 40.0 | 15 | 0 | 70 | 15 | |

| Analyte | mg/L |
|-----------------|------|
| 1. Fluoride | 2 |
| 2. Acetate | 10 |
| 3. Glycolate | 10 |
| 4. Propionate | 10 |
| 5. Formate | 10 |
| 6. Chloride | 5 |
| 7. Nitrite | 10 |
| 8. Bromide | 10 |
| 9. Nitrate | 10 |
| 10. Carbonate | 30 |
| 11. Sulfite | 10 |
| 12. Sulfate | 10 |
| 13. Oxalate | 10 |
| 14. Phosphate | 15 |
| 15. Thiosulfate | 15 |
| 16. Iodide | 30 |
| 17. Thiocyanate | 30 |

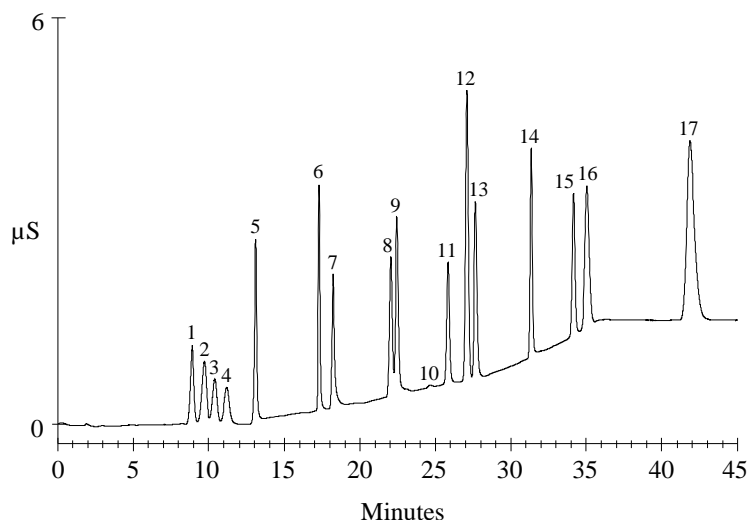


Figure 17
Gradient Elution of Organic Acid Anions, Common Inorganic Anions, and Hydrophobic Inorganic Anions

5.13. Gradient Analysis using Large Loop Injection

Figure 18 illustrates gradient analysis of low molecular weight organic acids and inorganic acids using large loop injection. A sodium hydroxide gradient is optimized by using a 2-mm Dionex IonPac AS11-HC column, which allows sulfate to resolve from carbonate so that carbonate does not interfere with the quantification of sulfate.

| | |
|-----------------------------------|--|
| Trap Column: | Dionex ATC-3, (Located between pump and injection valve) |
| Sample Volume: | 1.0 mL |
| Column: | Dionex IonPac AS11-HC Analytical and Dionex IonPac AG11-HC Guard |
| Eluent: | E1: Deionized water E2: 5.0 mM NaOH E3: 100 mM NaOH |
| Eluent Flow Rate: | 0.5 mL/min (2-mm), 2.0 mL/min (4-mm) |
| Operating Temperature: | 30°C |
| SRS Suppressor: | Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm) AutoSuppression External Water Mode |
| or MMS Suppressor: | Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm) |
| MMS Regenerant: | 50 mN H ₂ SO ₄ |
| Expected Background Conductivity: | 1.0 mM NaOH: 1 µS 50 mM NaOH: < 3.5 µS |
| Typical Operating Back Pressure: | 2700 psi (18.60 MPa) |

Gradient Conditions

| TIME (min) | %E1 | %E2 | %E3 | %E4 | Comments |
|----------------------|-----|-----|-----|-----|-------------------------------|
| Equilibration | | | | | |
| 0 | 80 | 20 | 0 | 0 | 1.0 mM NaOH for 7 min |
| 8.0 | 80 | 20 | 0 | 0 | |
| Analysis | | | | | |
| 7.1 | 80 | 20 | 0 | 0 | 1.0 mM NaOH, Inject |
| 13.0 | 80 | 20 | 0 | 0 | Inject Valve to Load Position |
| 14.0 | 95 | 0 | 5 | 0 | End isocratic analysis |
| 18.0 | 88 | 0 | 12 | 0 | Gradient analysis |
| 23.0 | 86 | 0 | 14 | 0 | |
| 27.0 | 77 | 0 | 23 | 0 | |
| 32.0 | 55 | 0 | 45 | 0 | |

| Analyte | mg/L |
|---------------|------|
| 1. Fluoride | 1 |
| 2. Acetate | 3 |
| 3. Formate | 1 |
| 4. Chloride | 1 |
| 5. Nitrite | 1 |
| 6. Bromide | 2 |
| 7. Nitrate | 2 |
| 8. Carbonate | - |
| 9. Sulfate | 1 |
| 10. Oxalate | 1 |
| 11. Phosphate | 3 |

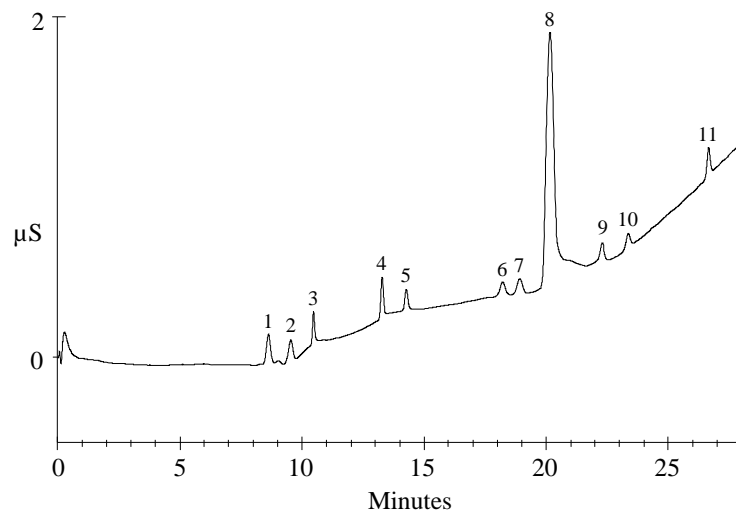


Figure 18
Gradient Analysis using Large Loop Injection

5.14. Clean-up After Humic Acid Samples

Solvent compatibility of the Dionex IonPac AS11-HC permits the use of organic solvents to effectively remove organic contaminants from the column. A Dionex IonPac AS11-HC column, after losing over 47% of its original capacity due to fouling with humic acid samples, can easily be restored to original performance by cleaning for 3 hours with 200 mM HCl in 80% tetrahydrofuran (THF).

Sample Loop Volume: 10 μ L
 Column: Dionex IonPac AS11-HC Analytical
 Eluent: 30 mM NaOH
 Eluent Flow Rate: 1.5 mL/min
 Operating Temperature: 30°C
 SRS Suppressor: Dionex Anion Self-Regenerating Suppressor, Dionex ASRS 300 (2-mm or 4-mm)
 AutoSuppression External Water Mode
 or MMS Suppressor: Dionex Anion MicroMembrane Suppressor, Dionex AMMS 300 (2-mm or 4-mm)
 MMS Regenerant: 50 mN H₂SO₄
 Expected Background Conductivity: < 3.5 μ S
 Long-term Storage Solution (> 1 week): 100 mM Sodium Borate
 Short-term Storage Solution (< 1 week): Eluent

| Analyte | mg/L |
|--------------|------|
| 1. Fluoride | 2.0 |
| 2. Chloride | 5.0 |
| 3. Nitrite | 10.0 |
| 4. Sulfate | 10.0 |
| 5. Bromide | 20.0 |
| 6. Nitrate | 20.0 |
| 7. Phosphate | 30.0 |

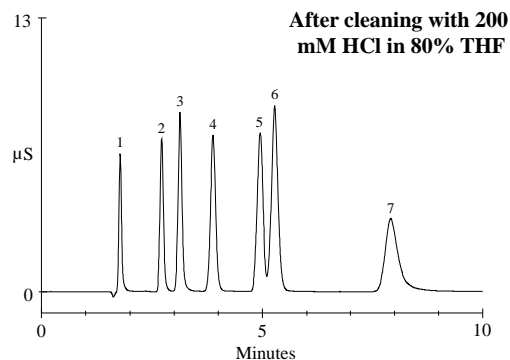
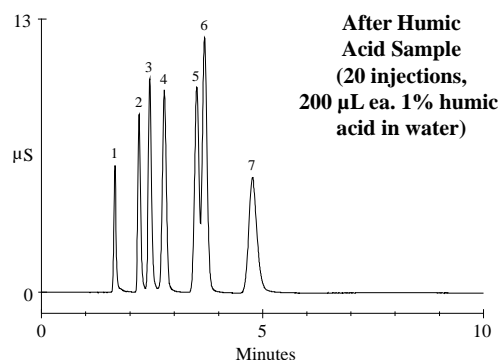
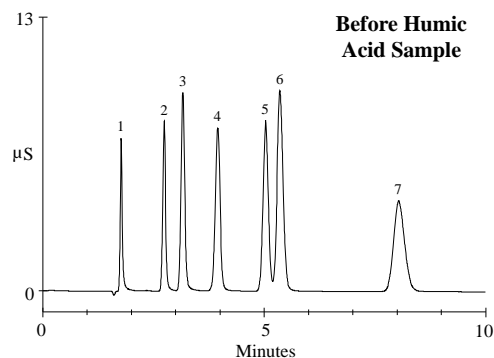


Figure 19
Clean-up after Humic Acid Samples

SECTION 6 – TROUBLESHOOTING GUIDE

The purpose of the Troubleshooting Guide is to help you solve operating problems that may arise while using Dionex IonPac AS11-HC 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. *For assistance, contact Technical Support for Dionex Products. In the U.S., call 1-800-346-6390. Outside the U.S., call the nearest Thermo Fisher Scientific office.*

Table 7
Dionex IonPac AS11-HC/Dionex IonPac AG11-HC Troubleshooting Summary

| Observation | Cause | Action | Reference Section |
|-------------------------------------|--|--|---------------------------|
| High Back Pressure | Unknown | Isolate Blocked Component | 6.1.1 |
| | Plugged Column Bed Supports | Replace Bed Supports, Filter Eluents, and Filter Samples | 6.1.2 |
| | Other System Components | Unplug, Replace | Component Manual |
| High Background Conductivity | Contaminated Eluents | Remake Eluents | 6.2, 6.2.1 |
| | Contaminated Trap Column | Clean Trap Column | 6.2.2 |
| | Contaminated Guard or Analyte Column | Clean Guard and Analytical/Capillary Column | 6.2.3 |
| | Contaminated Suppressor | Clean Suppressor | 6.2.4, Component Manual |
| | Contaminated Hardware | Clean Component | Component Manual |
| Poor Resolution | Poor Efficiency Due to Large System Void Volumes | Replumb System | 6.3.1.A, Component Manual |
| | Column Headspace | Replace Column | 6.3.1.B |
| Short Retention Times | Flow Rate Too fast | Recalibrate Pump | 6.3.2.A |
| | Conc. Incorrect Eluents | Remake Eluents | 6.3.2.B |
| | Column Contamination | Clean Column | 6.3.2.C, 6.3.2.D |
| Poor Front End Resolution | Conc. Incorrect 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 |
| | 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 Dionex IonPac AG11-HC (4-mm) Guard Column plus the Dionex IonPac AS11-HC (4-mm) Analytical Column when using the test chromatogram conditions should be equal or less than 2,250 psi. If the system pressure is higher than 2,250 psi, it is advisable to determine the cause of the high system pressure. The system should be operated with a Thermo Scientific Dionex 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/Capillary columns are connected (see Table 8, "Typical Dionex IonPac AS11-HC/Dionex IonPac AG11-HC Operating Back Pressures").

The Dionex Anion Self-Regenerating Suppressor 300 with backpressure loops 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 8
Typical Dionex IonPac AS11-HC/Dionex IonPac AG11-HC Operating Back Pressures

| Column | Typical Back Pressure with Aqueous eluents psi (MPa) at 30°C | Typical Back Pressure with Eluents containing Methanol (15%) psi (MPa) at 30°C | Flow Rate mL/min |
|---|--|--|------------------|
| Dionex IonPac AS11-HC 4-mm Analytical | 1900 (13.09) | 2450 (16.88) | 1.5 |
| Dionex IonPac AG11-HC 4-mm Guard | 150 (1.03) | 250 (1.72) | 1.5 |
| Dionex IonPac AS11-HC + Dionex IonPac AG11-HC 4-mm columns | 2050 (14.12) | 2700 (18.60) | 1.5 |
| Dionex IonPac AS11-HC 2-mm Analytical | 1800 (12.40) | 2450 (16.88) | 0.38 |
| Dionex IonPac AG11-HC 2-mm Guard | 150 (1.03) | 250 (1.72) | 0.38 |
| Dionex IonPac AS11-HC + Dionex IonPac AG11-HC 2-mm columns | 1950 (13.43) | 2700 (18.60) | 0.38 |
| Dionex IonPac AS11-HC 0.4 mm Capillary | 1500 (13.79) | 2000 | 0.015 |
| Dionex IonPac AG11-HC 0.4-mm Capillary Guard | 150 (1.38) | 250 | .0015 |
| Dionex IonPac AS11-HC + Dionex IonPac AG11-HC 0.4 mm columns | 1700 (15.17) | 2300 | .0015 |

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.

Table 9
Product Information

| Product | Dionex IonPac AS11-HC 4-mm Columns (P/N) | Dionex IonPac AS11-HC 2-mm Columns (P/N) | Dionex IonPac AS11-HC 0.4-mm Columns (P/N) |
|----------------------|---|---|---|
| Analytical Column | 052960 | 052961 | 078429 |
| Guard Column | 052962 | 052963 | 078430 |
| Bed Support Assembly | 042955 | 044689 | N/A |
| End Fitting | 052809 | 043278 | N/A |



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 finger-tight, 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.

6.1.3. Filter Eluent

Eluents containing particulate material or bacteria may clog the column inlet bed support. Filter water used for eluents through a 0.45 µm filter.

6.1.4. Filter Samples

Samples containing particulate material may clog the column inlet bed support. Filter samples through a 0.45 µm filter prior to injection.

6.2. High Background

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

Table 10
Background Conductivity

| Eluent | Expected Background Conductivity |
|-----------------------------------|----------------------------------|
| 1.0 mM NaOH | 0.5 - 1.0 μ S |
| 60 mM NaOH | 2.5 - 3.5 μ S |
| 60 mM NaOH/15% CH ₃ OH | 2.0 - 3.0 μ S |

6.2.1. Preparation of Eluents

- Make sure that the eluents and the regenerant are made correctly.
- Make sure that the eluents are made from chemicals with the recommended purity.
- 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 Trap Column

High background may be caused by contamination of the Dionex ATC-HC or Dionex ATC-3 with carbonate or other anions from the eluent.

- Clean the Dionex ATC-HC or Dionex ATC-3 (4 mm) with 100 mL of 2.0 M NaOH or 50 mL for the Dionex ATC-3 (2 mm).
- Rinse the Dionex ATC-HC or Dionex ATC-3 (4 mm) immediately with 20 mL of eluent or 10 mL of eluent for the Dionex ATC-3 (2 mm) into a beaker prior to use.

6.2.3. Contaminated Dionex CR-ATC Column

- Install a Dionex CR-ATC Anion Trap Column (P/N 060488 or 072078) if using a Dionex Eluent Generator with Dionex EGC KOH cartridge.
- If the Dionex CR-ATC becomes contaminated, please refer to Section 6, Clean-Up, in the Dionex CR-ATC Product Manual (Document No. 031910).

6.2.4. A Contaminated Guard or Analytical/Capillary Column

- Remove the Dionex IonPac AG11-HC Guard and Dionex IonPac AS11-HC Analytical/Capillary Columns from the system.
- Install a back pressure coil that generates approximately 1,500 psi and continue to pump eluent. If the background conductivity decreases, the column(s) is (are) the cause of the high background conductivity.
- To eliminate downtime, clean or replace the Dionex IonPac AG11-HC at the first sign of column performance degradation. Clean the column as instructed in, "Column Cleanup" (See Appendix A "Column Care").

6.2.5. Contaminated Hardware

Eliminate the hardware as the source of the high background conductivity.

- Bypass the columns and the suppressor.
- Install a back pressure coil that generates approximately 1,500 psi and continue to pump eluent.
- Pump deionized water with a specific resistance of 18.2 megohm-cm through the system.
- The background conductivity should be less than 2 μ S. If it is not, check the detector/conductivity cell calibration by injecting deionized water directly into it. See the appropriate manual for details.

6.2.6. A Contaminated Dionex ASRS 300, Dionex ACES 300 or Dionex AMMS 300 Suppressor

If the above items have been checked and the problem persists, the Dionex Anion Self-Regenerating Suppressor, the Dionex Anion Capillary Electrolytic Suppressor or the Dionex Anion MicroMembrane Suppressor is probably causing the problem. For details on Dionex Anion Self-Regenerating Suppressor operation, refer to the Dionex Anion Self-Regenerating Suppressor 300 Product Manual (Document No. 031956). For details on Dionex Anion Membrane Suppressor 300 operation, refer to the Product Manual (Document No. 031727) for assistance. For details on the Dionex Anion Capillary Electrolytic Suppressor 300 (Dionex ACES 300) operation, refer to the product manual (Document No. 065388) for assistance.

- A. Check the power level and alarms on the Dionex SRS Control.
- B. Check the regenerant flow rate at the REGEN OUT port of the Dionex ASRS if operating in the AutoSuppression External Waster mode or the Chemical Suppression mode or the Dionex AMMS.
- C. Check the eluent flow rate.
- D. If you are using a Dionex AutoRegen accessory with the Dionex ASRS in the Chemical Suppression Mode or the Dionex AMMS, prepare fresh regenerant solution.
- E. Test both the suppressor and the Dionex AutoRegen Regenerant Cartridge for contamination. If the background conductivity is high after preparing fresh regenerant and bypassing the Dionex AutoRegen Regenerant Cartridge, you probably need to clean or replace your Dionex ASRS, Dionex ACES or Dionex AMMS.

If the background conductivity is low when freshly prepared regenerant is run through the Dionex ASRS or Dionex AMMS without an AutoRegen accessory in-line, test the Dionex AutoRegen Regenerant Cartridge to see if it is expended.

- A. Connect the freshly prepared regenerant to the Dionex AutoRegen Regenerant Cartridge.
- B. Pump approximately 200 mL of regenerant through the Dionex AutoRegen Regenerant Cartridge to waste before recycling the regenerant back to the regenerant reservoir. If the background conductivity is high after placing the Dionex AutoRegen accessory in-line, you probably need to replace the Dionex AutoRegen Regenerant Cartridge. Refer to the “Dionex AutoRegen Regenerant Cartridge Refill Product Manual” (Document No. 032852) for assistance.

6.3. Poor Peak Resolution

One of the unique features of the Dionex IonPac AS11-HC is fast equilibration time in gradient applications from the last eluent (high ionic strength) to the first eluent (low ionic strength). The actual equilibration time depends on the ratio of the strongest eluent concentration to the weakest eluent concentration. Typically equilibration times range from 7 to 10 minutes.

If increased separation is needed for the first group of peaks, dilute eluent E1. This part of the chromatogram is run isocratically with E1.

Due to different system configurations, the gradient profile may not match the gradient shown in the example. The gradient conditions can be adjusted to improve resolution or to adjust retention times either by changing the gradient timing or by changing the gradient eluent proportions.

- A. Keep the concentrations of E1 and E2 constant and adjust the gradient time. This is the simplest way to compensate for total system differences if resolution is the problem.
- B. Change the proportions of E1 and E2 and adjust the gradient time. This approach requires more time to develop and more knowledge in methods development work. Its advantage is that it allows a method to be tailored for a particular application, where selectivity, resolution, and total run time are optimized. Be aware Poor peak resolution can be due to 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



NOTE

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.



NOTE

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 Appendix A "Column Care", for recommended column cleanup procedures.

**NOTE**

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, Appendix A, "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.

**NOTE**

For assistance, contact Technical Support for Dionex Products. In the U.S., call 1-800-346-6390. Outside the U.S., call the nearest Thermo Fisher Scientific office.

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/capillary 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/capillary columns may be the problem. Swap components, one at a time, in the system prior to the analytical/capillary 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 Appendix A "Column Care".

**NOTE**

For assistance, contact Technical Support for Dionex Products. In the U.S., call 1-800-346-6390. Outside the U.S., call the nearest Thermo Fisher Scientific office.

- 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.

6.3.5. Poor Efficiency Using Capillary Columns

Incorrectly installed fittings on capillary tubing can increase void volumes, causing chromatograms with tailing peaks.

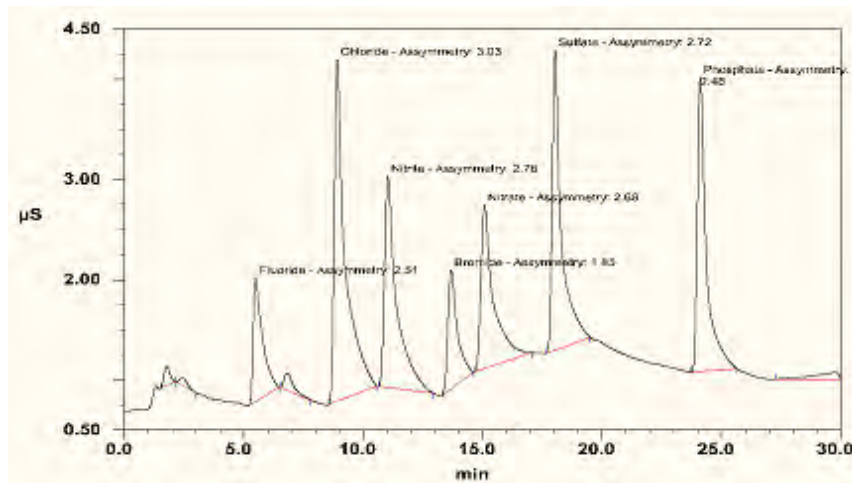


Figure 20
Tailing Peaks Caused by Incorrectly Installed
Capillary Tubing Fittings

When connecting a capillary tube fitting, make sure that the ferrule and fitting bolt are at least 2 mm (0.1 in) from the end of the tubing before you insert the tubing into the port. Do not place the ferrule and fitting bolt flush with the end of the tubing. Figure 21 illustrates the correct and incorrect placement of the ferrule and fitting bolt on the tubing.

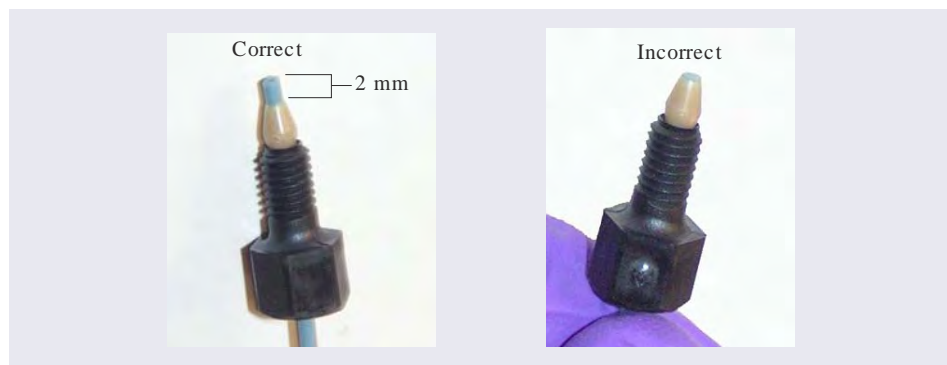


Figure 21
Correct and Incorrect Ferrule and
Fitting Bolt Placement for Capillary Tubing Connections

APPENDIX A – COLUMN CARE

A.1 Recommended Operation Pressures

Operating a column above its recommended pressure limit can cause irreversible loss of column performance. The maximum recommended operating pressure for Dionex IonPac AS11-HC columns is 4,000 psi (27.57 MPa).

A.2 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.

A.3 Column Storage

For short-term storage (< 1 week), use Eluent, for long-term storage (> 1 week), use 100 mM Sodium Borate for the column storage solution. Flush the column for a minimum of 10 minutes with the storage solution. Cap both ends securely, using the plugs supplied with the column.

A.4 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; however, the following precautions should be observed.



WARNING

- *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 < 5% levels and the ionic strength of the eluent to < 50 mM levels to avoid creating high pressure zones in the column that may disrupt the uniformity of the column packing.

A.4.1 Choosing the Appropriate Cleanup Solution

| Contamination | Solution |
|---|--|
| Hydrophilic Contamination of Low Valence | Concentrated hydroxide solutions such as a 10X concentrate of the most concentrated eluent used in the application is sufficient to remove hydrophilic contamination of low valence. |
| High Valence Hydrophilic Ions Contamination | Concentrated acid solutions such as 1 to 3 M HCl will remove high valence hydrophilic ions by ion suppression and elution by the chloride ion. |
| Metal Contamination | Metal contamination often results in asymmetric peak shapes and/or variable analyte recoveries. For example, iron or aluminum contamination often results in tailing of sulfate and phosphate. Aluminum contamination can also result in low phosphate recoveries. |
| | Concentrated acid solutions such as 1 to 3 M HCl remove a variety of metals. If after acid treatment, the chromatography still suggests metal contamination, treatment with chelating acids such as 0.2 M oxalic acid is recommended. |
| Nonionic and Hydrophobic Contamination | Organic solvents can be used alone if the contamination is nonionic and hydrophobic. The degree of nonpolar character of the solvent should be increased as the degree of hydrophobicity of the contamination within the range of acceptable solvents. |
| Ionic and Hydrophobic Contamination | Concentrated acid solutions such as 1 to 3 M HCl can be used with compatible organic solvents to remove contamination that is ionic and hydrophobic. The acid suppresses ionization and ion exchange interactions of the contamination with the resin. |
| | A frequently used cleanup solution is 200 mM HCl in 80% acetonitrile. This solution must be made immediately before use because the acetonitrile will decompose in the acid solution during long term storage. |

A.4.2 Column Cleanup Procedure

- A. Prepare a 500 mL solution of the appropriate cleanup solution using the guidelines in Section B.4.1, "Choosing the Appropriate Cleanup Solution."
- B. Disconnect the Dionex ASRS 300 or Dionex AMMS 300 from the Dionex IonPac AS11-HC Analytical/Capillary Column. If your system is configured with both a guard column and an analytical/capillary column, reverse the order of the guard and analytical/capillary 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/capillary column and a guard column in series, ensure that the guard column is placed after the analytical/capillary column in the eluent flow path. Contaminants that have accumulated on the guard column can be eluted onto the analytical/capillary column and irreversibly damage it. If in doubt, clean each column separately.

- C. Set the pump flow rate to 1.0 mL/min for a Dionex IonPac AS11-HC 4-mm Analytical or Guard Column or set the pump flow rate to 0.25 mL/min for a Dionex IonPac AS11-HC 2-mm Analytical or Guard Column. Set the pump flow rate to 0.010 mL/min for a Dionex IonPac AS11-HC Capillary or Capillary Guard Column.
- D. Rinse the column for 10 minutes with deionized water before pumping the chosen cleanup solution over the column.
- E. Pump the cleanup solution through the column for 60 minutes. Note in the case of Section B.4.1, "Choosing the Appropriate Cleanup Solution," Part B, "High valency hydrophilic ions," a step gradient is used for column cleanup.
- F. Rinse the column for 10 minutes with deionized water before pumping eluent over the column.
- G. Equilibrate the column(s) with eluent for at least 30 minutes before resuming normal operation.
- H. Reconnect the Dionex ASRS 300 or Dionex AMMS 300 to the Dionex IonPac AS11-HC Analytical/Capillary Column and place the guard column in line between the injection valve and the analytical/capillary column if your system was originally configured with a guard column.

APPENDIX B – SYSTEM CONFIGURATION

Table B1
Configuration

| CONFIGURATION | 2 mm | 4 mm | 0.4 mm |
|--|--|--|---|
| Eluent Flow Rate | 0.38 mL/min | 1.5 µL/min | 0.015 µL/min |
| SRS Suppressor | Dionex ASRS 300 (P/N 061562) | Dionex ASRS 300 (P/N 061561) | N/A |
| MMS Suppressor | Dionex AMMS 300 (P/N 056751) | Dionex AMMS 300 (P/N 056750) | N/A |
| ACES Suppressor | N/A | N/A | Dionex ACES 300 (P/N 072052) |
| NOTE: <i>Do not run suppressors over 40°C. If application requires a higher temperature, place suppressor outside of chromatographic oven.</i> | | | |
| Injection Loop | 2 - 15 µL Use the Rheodyne Microinjection Valve, Model No. 9126 P/N 044697) for full loop injections <15 µL. | 10 - 50 µL | 0.4 µL (typical) |
| System Void Volume | Eliminate switching valves, couplers and the Dionex GM-3 Gradient Mixer. Use only the 2 mm Dionex GM-4 Mixer (P/N 049135). | Minimize dead volumes. Switching valves, couplers can be used. Use the Dionex GM-2, GM-3 or recommended gradient mixers. | Use only in an IC system equipped for capillary analysis. |
| Pumps | Use the Dionex ICS 2100/5000, or Dionex GS50/GP50/GP40/IP20/IP25 in Microbore Configuration with a Microbore Dionex GM-4 (2 mm) Gradient Mixer. The Dionex 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 Dionex ICS 2100/5000, or Dionex GP40/GP50/IP20/IP25 in Standard-Bore Configuration. The Dionex GM-3 Gradient Mixer should be used for gradient analysis on systems other than the Dionex GP50. The Dionex GP40 has an active mixer. | Use only a pump designed for capillary flow rates such as the Dionex ICS-5000 capillary pump. |
| NOTE: <i>Use of a Dionex EGC-KOH cartridge (P/N 074532 or 072076 in conjunction with a Dionex CR-ATC P/N 060477 or 072078) for gradient applications is highly recommended for minimum baseline change when performing eluent step changes or gradients.</i> | | | |
| Chromatographic Module | A thermally controlled column oven such as the Dionex LC25,LC30,ICS-10,11,15,16,20,2100,3000,5000 DC | A thermally controlled column oven such as the Dionex LC25,LC30,ICS-10,11,15,16,20,2100,3000,5000 DC | A thermally controlled column compartment such as the Dionex ICS-5000 DC or Dionex IC-Cube. |

| CONFIGURATION | 2 mm | 4 mm | 0.4 mm |
|------------------|--|---|---|
| Detectors | <p>Dionex Conductivity Detector P/N 061830</p> <p>Dionex AD20/AD25 Cell (6 mm, 7.5 μL, P/N 046423)</p> <p>Dionex VDM-2 Cell (3 mm, 2.0 μL) (P/N 043120)</p> <p>Dionex CD20, CD25, CD25A, ED40, ED50, or ED50A</p> <p>Dionex Conductivity Cell with Dionex DS3 P/N 044130 or Dionex Conductivity Cell with shield P/N 044132</p> <p>Dionex CDM-2/CDM-3 Cell P/N 042770</p> <p>Replace the Dionex TS-1 with the Dionex TS-2 (P/N 043117) on the Dionex CDM-2 or the Dionex CDM-3. The Dionex TS-2 has been optimized for 2 mm operation. Do not use the Dionex TS-2 or the Dionex TS-1 with the Dionex ED40/ED50/ED50A or the Dionex CD20/CD25/CD25A.</p> <p>Ensure 30–40 psi back pressure.</p> | <p>Dionex Conductivity Detector P/N 061830</p> <p>Dionex AD20/AD25 Cell (10 mm, 9 μL, P/N 049393)</p> <p>Dionex VDM-2 Cell (6 mm, 10 μL) P/N 043113</p> <p>Dionex CD20, CD25, CD25A, ED40, ED50, or ED50A</p> <p>Dionex Conductivity Cell with Dionex DS3 P/N 044130 or Dionex Conductivity Cell with shield P/N 044132</p> <p>Dionex CDM-2/CDM-3 Cell P/N 042770</p> <p>Either the Dionex TS-1 with the Dionex TS-2 can be used with the Dionex CDM-2 or the Dionex CDM-3. Do not use the Dionex TS-2 or the Dionex TS-1 with the Dionex ED40/ED50/ED50A or the Dionex CD20/CD25/CD25A.</p> <p>Ensure 30–40 psi back pressure.</p> | <p>Use only a conductivity detector designed for capillary flow rates such as the Dionex ICS-5000 Capillary CD.</p> |

Table B2
Tubing Back Pressures

| Color | Part Number | I.D. inch | I.D. cm | Volume mL/ft | Back Pressure Psi/ft. at 1 mL/min | Back Pressure Psi/ft. at 0.25 mL/min | Back Pressure Psi/cm. at 1 mL/min |
|------------|-------------|-----------|---------|--------------|-----------------------------------|--------------------------------------|-----------------------------------|
| Green | 044777 | 0.030 | 0.076 | 0.137 | 0.086 | 0.021 | 0.003 |
| Orange | 042855 | 0.020 | 0.051 | 0.061 | 0.435 | 0.109 | 0.015 |
| Blue | 049714 | 0.013 | 0.033 | 0.026 | 2.437 | 0.609 | 0.081 |
| Black | 042690 | 0.010 | 0.025 | 0.015 | 6.960 | 1.740 | 0.232 |
| Red | 044221 | 0.005 | 0.013 | 0.004 | 111.360 | 27.840 | 3.712 |
| Yellow | 049715 | 0.003 | 0.008 | 0.001 | 859.259 | 214.815 | 28.642 |
| Light Blue | 071870 | 0.0025 | 0.006 | 0.0009 | 1766.0 | 441.0 | 58.0 |

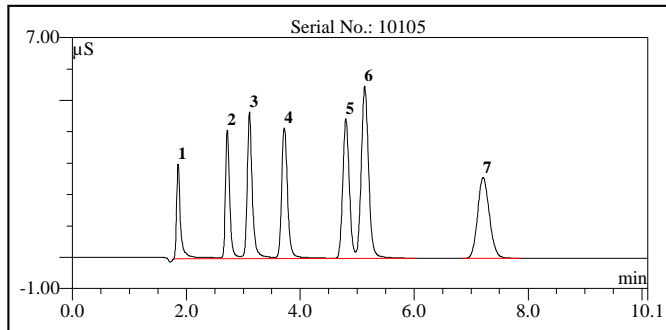
APPENDIX C – QUALITY ASSURANCE REPORT

C.1 Dionex IonPac AS11-HC 4-mm QAR

Dionex IonPac™ AS11-HC
Analytical (4 x 250 mm)
Product No. 052960

Date: 23-Jan-12 10:46
Serial No. : 010105
Lot No. : 011-15-159

Eluent: 30mM NaOH
Flow Rate: 1.50 mL/min
Temperature: 30 °C
Detection: Suppressed Conductivity
Suppressor: Anion Self-Regenerating Suppressor (ASRS™ 300 4mm)
 AutoSuppression™ Recycle Mode
Applied Current: 112 mA
Injection Volume: 10 µL
Storage Solution: 100 mM Sodium Tetraborate



| No. | Peak Name | Ret.Time (min) | Asymmetry (AIA) | Resolution (EP) | Efficiency (EP) | Concentration (mg/L) |
|-----|-----------|-------------------|--------------------|--------------------|--------------------|-------------------------|
| 1 | Fluoride | 1.85 | 2.1 | 7.44 | 4898 | 2.0 |
| 2 | Chloride | 2.72 | 1.4 | 2.82 | 7255 | 5.0 |
| 3 | Nitrite | 3.11 | 1.4 | 3.67 | 6920 | 10.0 |
| 4 | Sulfate | 3.72 | 1.3 | 5.56 | 6573 | 10.0 |
| 5 | Bromide | 4.80 | 1.2 | 1.53 | 8671 | 20.0 |
| 6 | Nitrate | 5.13 | 1.2 | 6.92 | 8120 | 20.0 |
| 7 | Phosphate | 7.21 | 1.2 | n.a. | 5876 | 30.0 |

QA Results:

| Analyte | Parameter | Specification | Results |
|---------|----------------|---------------|---------|
| Sulfate | Efficiency | >=4500 | Passed |
| Sulfate | Asymmetry | 1.0-1.8 | Passed |
| Sulfate | Retention Time | 3.36-4.35 | Passed |
| | Pressure | <=2200 | 1610 |

Production Reference:

Datasource: Column
 Directory: CPC\CPC_6
 Sequence: 1458315_AS11- HC_4mm_ZN
 Sample No.: 76

6.80 SR11 Build 3160 (183147) (Demo-Installation)

Chromeleon Dionex Corp. 1994-2012

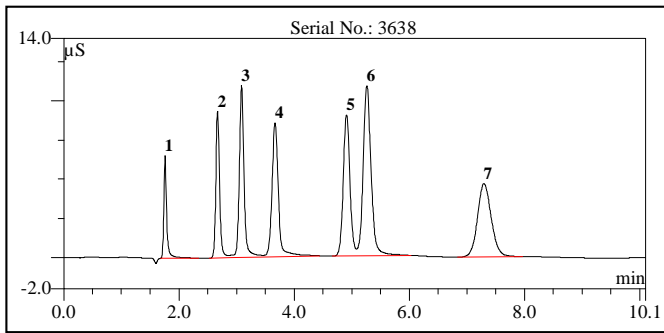
066758-05 (QAR)

C.2 Dionex IonPac AS11-HC 2-mm QAR

IonPac® AS11-HC
Analytical (2 x 250 mm)
Product No. 052961

Date: 06-Dec-11 07:48
Serial No. : 003638
Lot No. : 011-15-115

Eluent: 30 mM NaOH
Flow Rate: 0.38 mL/min
Temperature: 30 °C
Detection: Suppressed Conductivity
Suppressor: Anion Self-Regenerating Suppressor (ASRS® 300 2mm)
 AutoSuppression® Recycle Mode
Applied Current: 29 mA
Injection Volume: 2.5 µL
Storage Solution: 100 mM Sodium Tetraborate



| No. | Peak Name | Ret.Time (min) | Asymmetry (AIA) | Resolution (EP) | Efficiency (EP) | Concentration (mg/L) |
|-----|-----------|-------------------|--------------------|--------------------|--------------------|-------------------------|
| 1 | Fluoride | 1.76 | 1.5 | 9.28 | 7220 | 2.0 |
| 2 | Chloride | 2.67 | 1.2 | 3.28 | 8669 | 5.0 |
| 3 | Nitrite | 3.09 | 1.1 | 3.56 | 7787 | 10.0 |
| 4 | Sulfate | 3.67 | 1.1 | 6.27 | 6175 | 10.0 |
| 5 | Bromide | 4.91 | 1.1 | 1.55 | 8640 | 20.0 |
| 6 | Nitrate | 5.26 | 1.2 | 6.09 | 7602 | 20.0 |
| 7 | Phosphate | 7.29 | 1.2 | n.a. | 4647 | 30.0 |

QA Results:

| Analyte | Parameter | Specification | Results |
|---------|----------------|---------------|---------|
| Sulfate | Efficiency | >=4320 | Passed |
| Sulfate | Asymmetry | 1.0-1.8 | Passed |
| Sulfate | Retention Time | 3.36-4.35 | Passed |
| | Pressure | <=2200 | 1403 |

Production Reference:

Datasource: Column
 Directory CPF\CPF_5
 Sequence: 1429294_AS11HC_2mm_BR
 Sample No.: 52

6.80 SR11 Build 3160 (183147) (Demo-Installation)

Chromeleon® Dionex Corp. 1994-2011

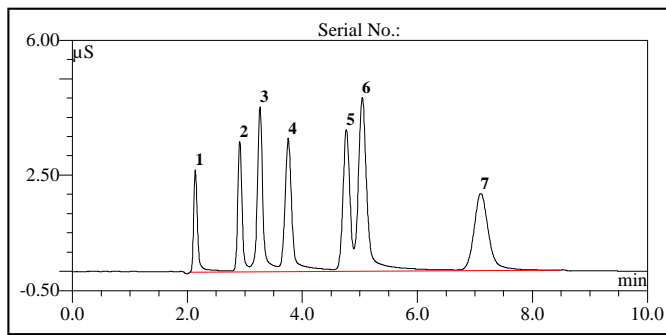
066756-04 (QAR)

C.3 Dionex IonPac AS11-HC 0.4-mm QAR

IonPac® AS11-HC
Analytical (0.4 x 250 mm)
Product No. 072489

Date: 22-Jan-12 14:09
Name : 000006
Lot No. : 011-038-015
Tubing ID (inch) 0.01580

Eluent: 30 mM NaOH
Flow Rate: .015 ml/min
Temperature: 30 °C
Detection: Suppressed Conductivity
Suppressor: Anion Capillary Electrolytic Suppressor (ACES®)
 AutoSuppression® Recycle Mode
Applied Current: 10.0
Injection Volume: 0.4 microliter
Storage Solution: 100 mM Sodium Tetraborate



| No. | Peak Name | Ret.Time (min) | Asymmetry (ATA) | Resolution (EP) | Efficiency (EP) | Concentration (mg/L) |
|-----|-----------|-------------------|--------------------|--------------------|--------------------|-------------------------|
| 1 | Fluoride | 2.14 | 1.4 | 6.49 | 6143 | 0.5 |
| 2 | Chloride | 2.91 | 1.2 | 2.47 | 8016 | 1.3 |
| 3 | Nitrite | 3.26 | 1.1 | 2.81 | 7110 | 2.5 |
| 4 | Sulfate | 3.75 | 1.1 | 4.88 | 5750 | 2.5 |
| 5 | Nitrate | 4.76 | n.a. | 1.20 | 7712 | 5.0 |
| 6 | Bromide | 5.04 | n.a. | 6.04 | 6709 | 5.0 |
| 7 | Phosphate | 7.10 | 1.2 | n.a. | 4206 | 7.5 |

QA Results:

| Analyte | Parameter | Specification | Results |
|---------|----------------|---------------|---------|
| Sulfate | Efficiency | >=4500 | Passed |
| Sulfate | Asymmetry | 1.0-1.8 | Passed |
| Sulfate | Retention Time | 3.36-4.35 | Passed |
| | Pressure | <=2200 | 1420 |

Production Reference:

Datasource: CapIC
 Directory Mjayaraman\AS11HC
 Sequence: AS11HC-CAP
 Sample No.: 324

6.80 SR10 Build 2818 (166959)

Chromeleon® Dionex Corp. 1994-2012

066756-03 (QAR)