Thermo Scientific

Acclaim™ Surfactant

Product Manual

P/N: 065087-03 February 2012

Part of Thermo Fisher Scientific

Thermo SCIENTIFIC

PRODUCT MANUAL

for the

ACCLAIM SURFACTANT GUARD COLUMN

4.3 x 10mm, P/N 063215
2.1 x 10mm, P/N 069693
3.0 x 10mm, P/N 071991
4.6 x 10mm, P/N 069701

ACCLAIM SURFACTANT ANALYTICAL COLUMN

5µm, 4.6 x 150mm, P/N 063201
5µm, 4.6 x 250mm, P/N 063203
5µm, 2.1 x 150mm, P/N 068123
3µm, 3.0 x 150mm, P/N 070084
3µm, 2.1 x 150mm, P/N 070085

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SECTION 1 – INTRODUCTION TO THE ACCLAIM SURFACTANT COLUMN

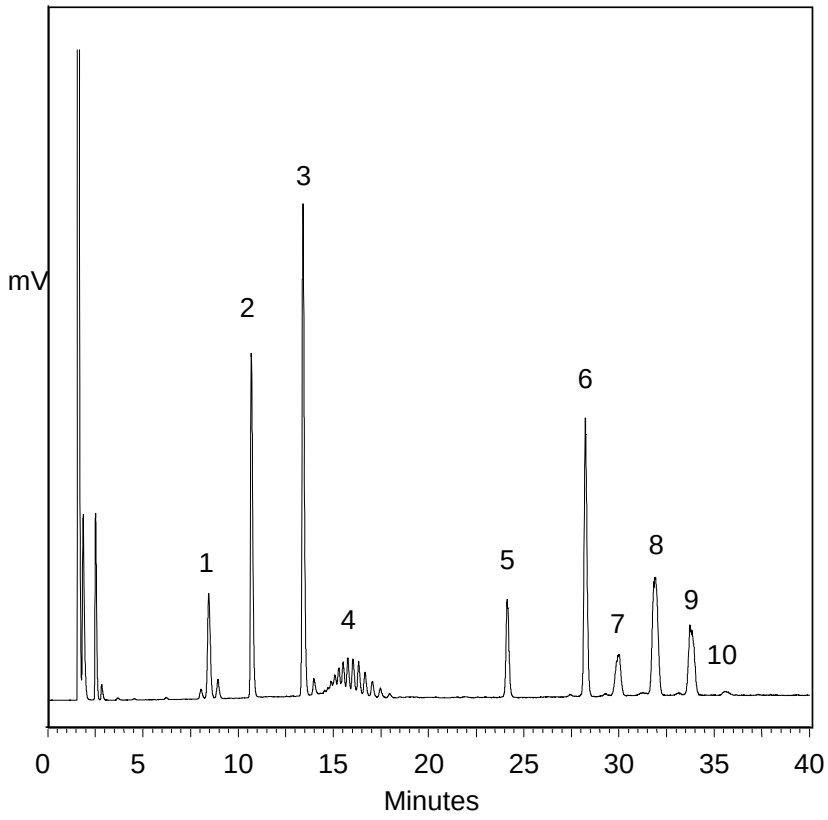
1.1 FEATURES OF THE ACCLAIM SURFACTANT COLUMN

The Acclaim Surfactant column is a specialty silica-based column that incorporates a proprietary bonding chemistry developed specifically for the separation of surfactants. The separation mechanism of the Acclaim Surfactant is a combination of reversed phase, anion-exchange, and dipole-dipole interactions. The Acclaim Surfactant column offers ideal selectivity and unprecedented capability for separating cationic, nonionic, and anionic surfactants in a single chromatographic run, using simple and volatile (MS compatible) mobile phases.

Acclaim Surfactant columns consist of 5-µm,or 3-µm, high purity spherical silica particles with 120 Å diameter pores bonded with proprietary functional groups, which provide unsurpassed resolution and peak symmetry for a variety of surfactants, as shown in Figure 1. The Acclaim Surfactant column provides several features:

- Ideal selectivity for simultaneous separation of anionic, nonionic and cationic surfactants
- Excellent peak shapes for cationic surfactants
- Improved resolution for oligomers of ethoxylated surfactants
- Compatibility of highly aqueous mobile phase conditions
- Proven chromatographic conditions suitable for various detection methods (ELSD, UV, conductivity detection, etc.)
- Broad range of applications

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile phase:	A –ACN, B- 0.1 M NH_4OAc, pH 5.4
Gradient:	25% to 85% A in 30 min., then hold for 10 min
Temperature:	30 °C
Flow rate:	1 mL/min
Injection vol.:	25 µL
Detection:	ELS Detector



Analytes:
1. Xylene sulfonate
2. Lauryldimethylbenzyl ammonium chloride
3. Octylphenoxyethoxyethyl dimethylbenzyl ammonium chloride
4. Triton X-100
5. Decyl sulfate
6. Dodecyl sulfate
7-10. Dodecylbenzene sulfonate

Figure 1
Separation of Cationic, Nonionic, and Anionic Surfactants

1.2 ACCLAIM SURFACTANT OPERATING LIMITS AND SPECIFICATIONS

Stationary Phase	Particle size	Column Dimensions	P/N	Max Recommended Pressure	Typical Flow Rate
Surfactant	3 µm	2.1x150 mm	070085	5500 psi	0.2 - 0.5 mL/min
		3.0x150 mm	070084	5500 psi	0.4 - 1.0 mL/min
	5 µm	2.1x150 mm	066866	5000 psi	0.2 - 0.5 mL/min
		4.6x150 mm	063201	5500 psi	0.8 - 2.0 mL/min
		4.6x250 mm	063203	5500 psi	0.8 - 2.0 mL/min

Shipping Solution: Acetonitrile / 0.1% Acetic Acid v/v 80/20
Storage Solution: Acetonitrile / 0.1% Acetic Acid v/v 80/20
Buffer pH Range: pH 2.5–7.5
Solvents: Acetonitrile, Methanol
Temperature Range: <60 °C

Physical
Bonding: Proprietary
Endcapping: Yes
% C: 12
Pore size: 120 Å
Surface area: 300 m^2/g
Particle size: 5 µm

1.3 FORMATS OF THE ACCLAIM SURFACTANT COLUMN

Acclaim Surfactant	Particle size	Column Dimensions	P/N	
Analytical	3 µm	2.1x150 mm	070085	
		3.0x150 mm	070084	
	5 µm	2.1x150 mm	068123	
		4.6x150 mm	063201	
		4.6x250 mm	063203	
Guard	5µm	4.3 x 10mm	063215	Requires Holder P/N 059456
		2.1 x 10mm	069693	Requires Holder V-2 P/N 069580
		3.0 x 10mm	071991	Requires Holder V-2 P/N 069580
		4.6 x 10mm	069701	Requires Holder V-2 P/N 069580

1.4 ACCLAIM SURFACTANT OPERATING CONDITIONS

The recommended pH range for Acclaim Surfactant columns is between pH 2.5 – 7.5. They are compatible with mobile phases containing 0–100% HPLC solvents, such as methanol or acetonitrile. The Acclaim Surfactant can be operated at any flow rate, as long as the backpressure remains below its recommended maximum pressure (see section 1.2). When setting up the analytical system, please be sure to check the special precautions listed in Section 3, "Installation".

1.5 KEY APPLICATIONS OF THE ACCLAIM SURFACTANT COLUMN

The Acclaim Surfactant column is a valuable tool for the analysis of all types of sample matrix, including food and beverages, pharmaceuticals, chemicals, and environmental samples.

Applications by Class of Compounds:

- Anionic surfactants
- Cationic surfactants
- Non-ionic surfactants
- Amphoteric surfactants

Application by Industries:

- Food and beverages
- Pharmaceuticals
- Chemicals
- Semiconductors
- Environmental

SECTION 2 – CHROMATOGRAPHY SYSTEM

Typical Flow Rate:	Please refer to Section 1.2
Injection Volume:	5–50 µL for the 4.6mm ID, 2-20 µL for the 3.0mm ID, and 1-10 µL for the 2.1mm ID
System Void Volume:	Minimize the lengths of all connecting tubing and remove all unnecessary switching valves and couplers.
Pumps:	Any HPLC pump capable of delivering gradients
Detectors:	UV, Conductivity, ELSD, MS, RI

2.1 DETECTORS

The Acclaim surfactant column is compatible with a variety of detectors and UV, ELSD and RI detectors. Example applications using each of these detectors are shown in Section 6 "Applications".

2.1.1 Evaporative Light Scattering Detection (ELSD)

When different surfactants are present in the sample with concentrations of no less than 50 ppm for each individual surfactant, ELSD is a good option. ELSD is a universal detection method that is compatible with gradient methods and is far more sensitive than RI. In addition, methods developed with ELSD can be easily transferred to LC-ESI-MS applications with little or no modification, because both detectors share the same mobile phase requirements. A UV detector can serve as a complementary tool and, if chosen, should be placed between the analytical column and ELSD.

2.1.2 Suppressed Conductivity Detection (SCD)

Suppressed conductivity detection can also be used for surfactant analysis and provides certain advantages for analyzing trace levels ionic surfactants in complex matrices. In fact, when analyzing ionic surfactants, especially at low concentrations (below 10 ppm), conductivity detection using a Dionex PEEK system, equipped with a gradient pump, electrochemical detector, MMS suppressor, and conductivity cell, is the best approach. Eluents for the separation of surfactants using conductivity detection include borate buffer and acetic acid. If required, a UV detector can serve as a complementary tool that should be placed between the analytical column and the MMS suppressor.

2.1.3 UV Detection

UV absorbance is the most popular detection method in HPLC, due to its ease of use and sensitivity. The drawback with this approach is that the analyte must have a chromophore to be detected and many surfactants do not. The UV detector can be used alone or with other detectors.

2.1.4 Mass Spectrometric Detection (MS)

Mass spectrometry (MS) is an inherently sensitive and universal method and has become the widely accepted tool for characterization of organic compounds. The soft ionization techniques, such as electrospray ionization (ESI), have greatly increased the applicability of MS detection to surfactant analysis. MS detectors are the detector of choice when less than 1 ppm individual surfactant is present and/or identification is required. Mass spectrometry compatible methods using ammonium acetate and acetonitrile have been developed. They are shown in Section 6, "Applications" in this manual.

MS detection can be used in series with other detectors, such as UV. Place the UV detector between the column and the MS or ELSD.

2.1.5 Refractive Index Detection (RI)

Although refractive index (RI) detection is a universal detection method capable of detecting all analytes, it is incompatible with gradient methods, exhibits low sensitivity, and thus is typically only used when other detection methods are not applicable or available.

2.2 PUMPS

Any HPLC pump can be used for the separation of surfactants with the Acclaim surfactant column. However, if suppressed conductivity detection is chosen, a PEEK pump from Dionex is recommended.

2.3 INJECTORS

Either an autosampler or a manual injector can be used for sample injection.

SECTION 3 - INSTALLATION

3.1 SYSTEM REQUIRMENTS

The Acclaim Surfactant column can be run on any HPLC system, including Dionex IC systems, and the UltiMate 3000 HPLC systems. Each of the possible configurations offers multiple sampling options. However, consistently reproducible quantification and an absence of disturbing artifacts are best achieved using the "full loop" mode and in conjunction with a 5 - 50 µL loop for the 4.6mm ID column, 2 - 20µL for the 3.0mm ID column, and 1 - 10µL for the 2.1mm ID column. Optimal reproducibility of retention time results can be achieved by regulating the temperature of the column using a column heater.

3.2 SYSTEM VOID VOLUME

For best performance, minimize the lengths of all connecting tubing and remove all unnecessary switching valves and couplers.

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.

3.3 THE INJECTION LOOP

DIONEX recommends using 5-50 µL injection loops for 4.6mm ID columns, 2 - 20µL for the 3.0mm ID columns, and 1-10 µL for 2.1mm ID columns.

3.4 COLUMN INSTALLATION

Connect the column with the direction arrow on the label pointing toward the detector.

3.5 COLUMN GUARD

When using a guard cartridge, install the cartridge in the guard holder. Connect the guard holder to the analytical column using the column connector fitting.

SECTION 4 – OPERATION

4.1 MOBILE PHASE SELECTION

4.1.1 Evaporative Light Scattering Detection (ELSD)

ELSD requires volatile buffers, such as ammonium acetate, ammonium formate, acetic acid, or formic acid. 20 to 100 mM buffer concentration is a good starting point. Acetonitrile is the preferred organic modifier because it generates a much lower backpressure on the column than does methanol.

4.1.2 Suppressed Conductivity Detection (SCD)

SCD requires a low conductivity or suppressible mobile phase to achieve high sensitivity. A good eluent for the analysis of cationic surfactants is 10 to 20 mM acetic acid with acetonitrile. For analysis of anionic surfactants, 10 to 30 mM ammonium borate buffer ~pH6 with acetonitrile is a good starting point. Note that when borate buffer is used, acetonitrile (not methanol) should be used as the organic modifier.

4.1.3 UV Detection

The mobile phase for the Acclaim Surfactant column with UV detection should have low UV background.

4.1.4 Mass Spectrometric Detection (MS)

Like ELSD, MS detection requires volatile buffers, such as ammonium acetate, ammonium formate, acetic acid, or formic acid. 20 to 100 mM buffer concentration is a good starting point. Acetonitrile is the preferred organic modifier because it generates a much lower backpressure on the column than does methanol.

4.1.5 Refractive Index Detection (RI)

The mobile phase for the Acclaim Surfactant column with RI detection should have low RI background.

4.2 VALIDATING COLUMN PERFORMANCE

Dionex recommends that you perform an efficiency test on your Acclaim Surfactant column before use. The purpose of column performance validation is to ensure no damage has occurred during shipping. Test the column using the conditions described on the Quality Assurance Report enclosed in the column box. Repeat the test periodically to track the column performance over time. Note that slight variations may be obtained on two different HPLC systems due to system electronic, plumbing, operating environment, reagent quality, column conditioning, and operator technique.

4.2.1 Procedure for Validating Column Performance:

1) Connect the column to the LC system.
2) Purge the column with the mobile phase listed on the QA report for 20 to 40 column volumes.
3) Inject the test mix shown in the QA report and collect the data.
4) Compare your result with the QA report provided in the column box.
5) If the chromatogram is satisfactory, you can use the column for your application work.

4.3 EQUILIBRATING THE COLUMN

Equilibrate the column after installing it for the first time. Always re-equilibrate the column prior to use following periods of storage.

Purge the column with the shipping or storage solvent until the baseline is stable. The shipping and recommended storage solvent is acetonitrile / 0.1% acetic acid v/v 80/20. Purge the column with acetonitrile / 0.1% acetic acid v/v 80/20. Then equilibrate the column with at least 15 column volumes of the mobile phase until a stable baseline is achieved.

4.4 QUICKSTART FOR ACCLAIM SURFACTANT COLUMN

4.4.1 Using the chromatographic conditions for Column Quality Assurance Report:

Column:	5 micron particle size
Mobile phase:	Acetonitrile/0.1M NH_4OAc, pH5.4 v/v 50/50
Flow rate:	4.6 x 150 mm use 1 mL/min ; 2.1 x 150mm use 0.2 mL/min
Temperature:	30 ˚C
Injection vol.:	5 µL
Detection:	UV at 220 nm

4.4.2 Column Quickstart

1) Prepare the mobile phase (see Section 4.7).
2) Install the mobile phase, 0.1% acetic acid, DI water, and acetonitrile on the LC pump.
3) Turn on the UV detector and set it to 220nm.
4) Prime the pump.
5) Install the column.
6) Flush the column with at least 15 column volumes of acetonitrile / 0.1% acetic acid v/v 80/20, then 10 column volumes of acetonitrile / 0.1% acetic acid v/v 50/50.
7) Equilibrate the column under initial mobile phase conditions for at least 15 minutes.
8) Conduct tests.
9) At the end of the day, flush the column with acetonitrile / 0.1% acetic acid v/v 50/50 for approximately 10 minutes.
10) Flush the column with acetonitrile / 0.1% acetic acid v/v 80/20 for approximately 10 minutes.
11) Turn off the pump and detector.
12) Remove the column and plug the ends, if the column will not be used for more than 2–3 days.

*For short-term storage (such as overnight), you may store the column in mobile phase.

4.5 CARING FOR THE COLUMN

To ensure the high performance of the Acclaim Surfactant column, the following guidelines should be followed:

1) Protect the column from contamination using an Acclaim Surfactant guard cartridge,
2) Make sure that solvents are miscible when changing mobile phases.
3) Always degas and filter mobile phases through a 0.22-µm membrane filter.
4) When switching to a new mobile phase, the column should be equilibrated for at least 30 column volumes before injecting the sample.
5) The recommended pH range is from pH 2.5 to 7.5. However, it is preferred that the column be used between pH3 and pH6.5 to achieve longer lifetime.
6) The column can be stored in mobile phase for short time storage (e.g. overnight). However, it is highly recommended that the column be stored in acetonitrile / 10-50 mM NH_4OAc buffer (pH 4.0-5.0) v/v 80/20 to 50/50 for long-term storage. If the mobile phase contained a buffer, first flush the column with 10 column volumes of acetonitrile / 10 mM NH_4OAc buffer (pH 5) v/v 50/50 before changing over to acetonitrile / 10-50 mM NH_4OAc buffer (pH 4.5) v/v 80/20. **Do not store the column under highly aqueous conditions for an extended period of time.**
7) The recommended operating maximum temperature is below 60 ˚C. In most cases, temperature control between ambient and 30 ˚C gives good results.
8) The recommended maximum back pressure is 4000 psi.

4.6 CHEMICAL PURITY REQUIREMENTS

Obtaining reliable, consistent and accurate results require mobile phases that are free of ionic and spectrophotometric impurities. Chemicals, solvents and de-ionized water used to prepare mobile phase should be of the highest purity available. Maintaining low trace impurities and low particle levels in mobile phases helps to protect your columns and system components. DIONEX cannot guarantee proper column performance when the quality of the chemicals, solvents and water used to prepare the mobile phase has been compromised.

4.6.1 Inorganic Chemicals

Reagent Grad 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 should be labeled, with the purity included on the lot analysis. Some recommended reagents are HPLC grade.

4.6.2 De-ionized Water

The de-ionized water used to prepare the mobile phase should be Type I Reagent Grade Water with a specific resistance of 18.2 megohm-cm. The de-ionized water should be free of ionized impurities, organics, microorganisms and particulate matter larger than 0.2 µm. Many commercial water purifiers are designed for HPLC applications and are suitable for these applications.

4.6.3 Solvents

The solvents used must be free from ionic and UV-absorbing 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 manufacture ultrahigh purity solvents that are compatible with HPLC and spectrophotometric applications. Use of these ultrahigh purity solvents will usually ensure that your chromatography is not affected by ionic impurities in the solvent. Currently at Dionex, we have obtained consistent results using High Purity Solvents manufactured by Burdick and Jackson and Optima® Solvents by Fischer Scientific.

When using a solvent in an ionic mobile phase, column generated backpressure will depend on the solvent used, concentration of the solvent, the ionic strength of the buffer and the flow rate used. The column back pressure will vary as the composition or the water-methanol or water-acetonitrile mixture varies. The practical backpressure limit for the Acclaim Surfactant columns is 4,000 psi (27.57 MPa).

NOTE
Since aqueous-methanol mixtures result in much higher back-pressures, aqueous-acetonitrile mobile phases in any ratio are preferred and recommended.

4.7 MOBILE PHASE PREPARATION

4.7.1 0.1 M Ammonium Acetate Buffer (1 L)

Dissolve 7.8 g ammonium acetate (43,131-1, Aldrich) in 1000 g D.I. water. Adjust the pH to the final value using acetic acid or ammonium hydroxide.

4.7.2 20 mM Sodium Borate Buffer (1 L)

Combine 200 mL of 0.1N sodium hydroxide solution (31,948-1, Aldrich) and 800 mL D.I. water and mix thoroughly. Add 31 g boric acid (>99.5%, 15660, Fluka) and sonicate the mixture under vacuum for 5 minutes.

4.7.3 20 mM Acetic Acid Buffer (1 L)

Dissolve 1.2 g of acetic acid (9524-33, J.T. Baker) in 1000 g DI water followed by sonication under vacuum for 5 minutes.

4.7.4 Mobile Phases Containing Solvents

When mixing solvents with water, mix the solvent with water on a volume-to-volume basis. For example, if a procedure requires a mobile phase of 10% acetonitrile, prepare the mobile phase by adding 100 mL of acetonitrile to a mobile phase reservoir. Then add 900 mL of the aqueous portion (DI water or buffer) to the acetonitrile in the reservoir. Using this procedure to mix the solvent with the aqueous phase will ensure that a consistent true volume/volume mobile phase is obtained. Premixing water with solvent will minimize the possibility of out-gassing in the detector cell.

NOTE
Degas the aqueous component of the mobile phase and then add to the solvent component. Avoid excessive purging or degassing of mobile phases containing solvents, if possible, since a volatile solvent can be 'boiled' off from the solution.

SECTION 5 – METHODS DEVELOPMENT

The Acclaim Surfactant column is compatible with a wide range of mobile phase systems that are compatible with UV, ELSD, MS, and conductivity detection. Not only can it be used as a QA column for the analysis of individual surfactants, but it is also valuable for the analysis of surfactants in complex sample matrices. In addition, by changing the composition of the mobile phase with respect to ionic strength or pH, the Acclaim Surfactant column can be used to identify the nature of species of interest in unknown sample, as shown in Figures 3 and 4.

NOTE

The Acclaim Surfactant column features reversed-phase anion-exchange and hydrogen-bonding interaction. Thus, it should not be used in the same way as conventional reversed-phase columns.

5.1 IONIC STRENGTH

The ionic strength is a powerful selectivity modifier for separating surfactants. Increasing the ionic strength of the mobile phase produces several results: a decrease in the retention time for anionic surfactants, an increase in the retention for cationic surfactants, but no change for nonionic surfactants. In this way, a clear distinction between the different classes of surfactants present in the sample can be made, as shown in Figure 2.

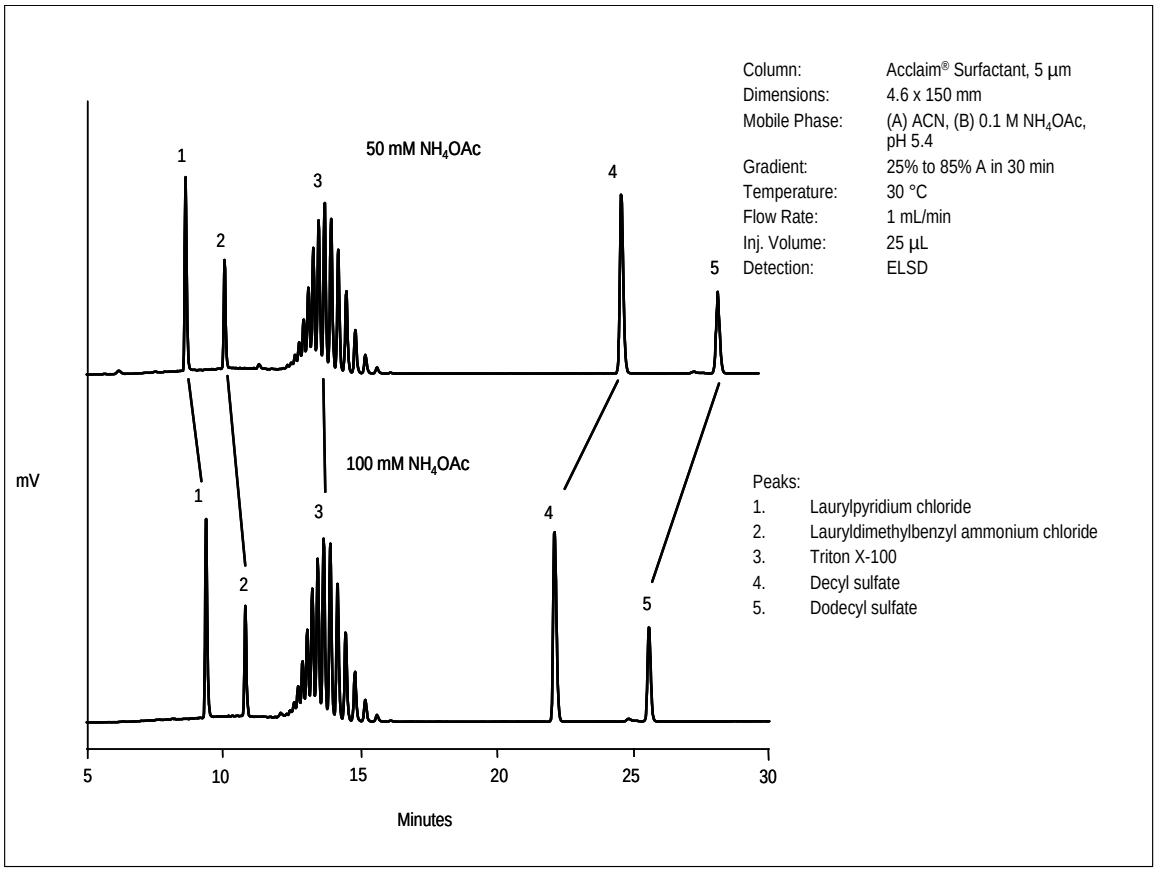


Figure 2
Effect of Ionic Strength on Selectivity

5.2 MOBILE PHASE pH

Another powerful selectivity modifier is the mobile phase pH. Increasing the pH of the mobile phase produces several results: a decrease in the retention time of anionic surfactants, an increase in the retention time of cationic surfactants, but no change in the retention time of non-ionic surfactants. These effects are shown in Figures 3 - 4.

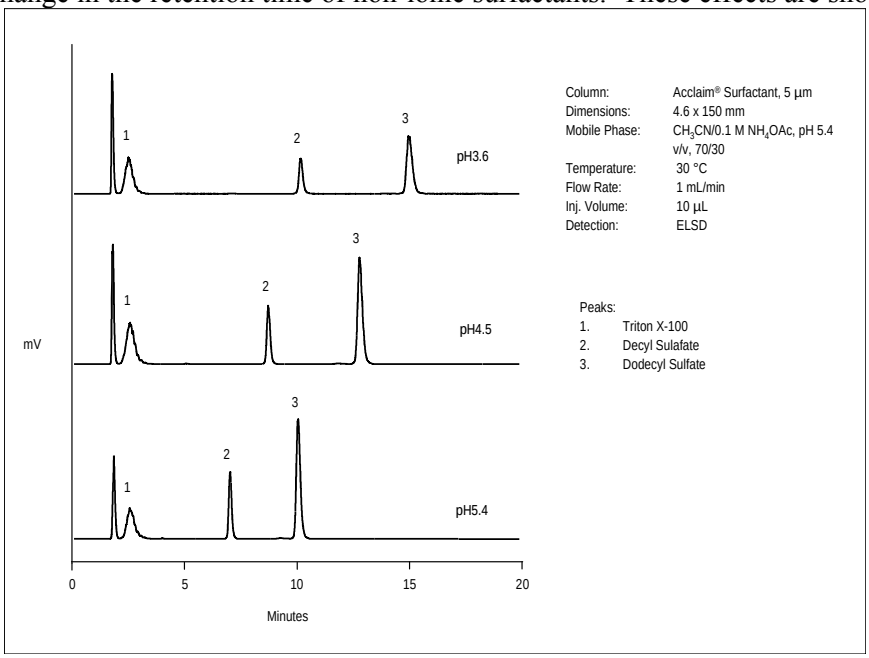


Figure 3
Effect of pH on the Selectivity of Anionic Surfactants

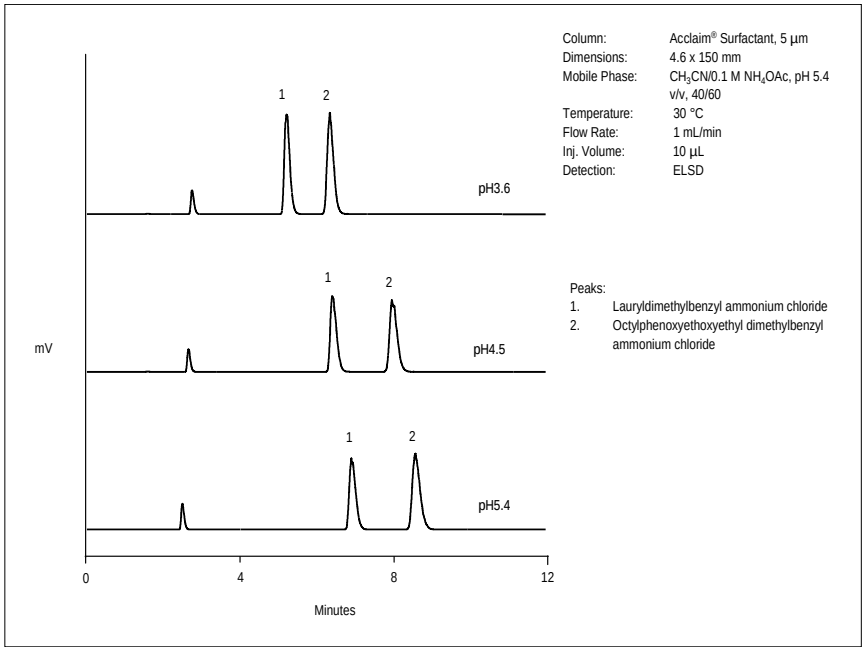


Figure 4
Effect of pH on the Selectivity of Cationic Surfactants

5.3 ORGANIC MODIFIER

Increasing the concentration of the organic modifier in the mobile phase generally results in a decrease in the retention time for all classes of surfactant. Acetonitrile or methanol is the recommended organic modifier. Acetonitrile is preferred over methanol because it generates less backpressure on the column, compared with methanol.

5.4 ION-PAIRING AGENT

The use of different ion-pairing agents affects the retention times of both cationic and anionic surfactants. For example, the use of tetramethyl ammonium acetate results in an increase in the retention time for anionic surfactants compared with the use of ammonium acetate. Similarly, the use of larger anions in the mobile phase gives rise to an increase in the retention time for cationic surfactants.

5.5 TEMPERATURE

Temperature has little effect on the selectivity of surfactants using the Acclaim Surfactant column; all the surfactants are eluted earlier. Since there is no advantage to higher temperatures, it is recommended that the Acclaim Surfactant column be used in a controlled environment, between 25 ^{o}C and 35 ^{o}C to ensure consistent retention times. Operation above 50 ^{o}C will result in an increased background signal using ELSD or MS detection.

SECTION 6 – APPLICATIONS

6.1 ANIONIC SURFACTANTS

6.1.1 Linear Alkylbenzene Sulfonates

Column: See chromatograms
Dimensions: 4.6 x 150 mm
Temperature: 30 °C
Flow rate: 1 mL/min
Injection vol.: 10 µL
Detection: UV, 225 nm

Mobile phases:
For Acclaim Surfactant
- ACN/100 mM NH_4OAc, pH 5.4 v/v 70/30

For Acclaim PA
- ACN/100 mM NH_4OAc, pH 5.4 v/v 50/50

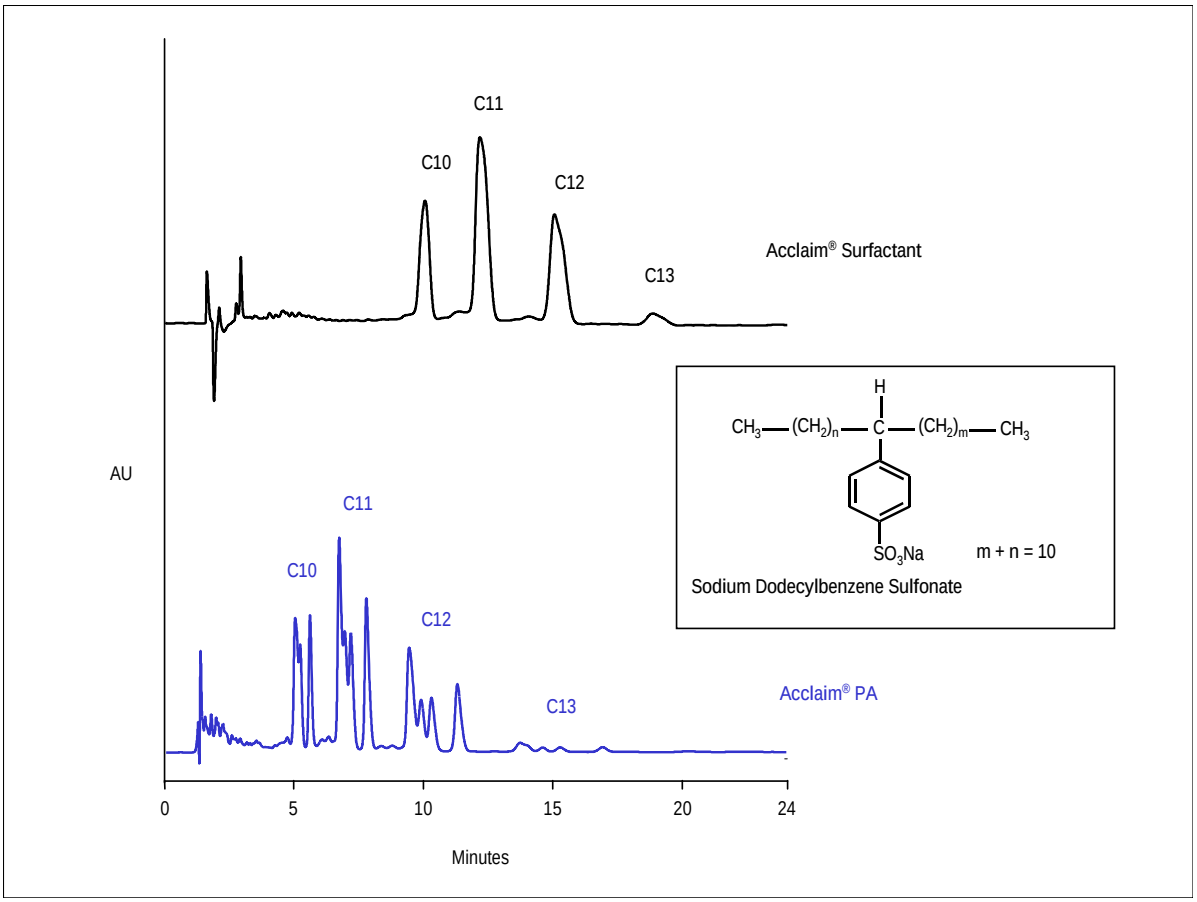


Figure 5
Separation of Linear Alkylbenzene Sulfonates

6.1.2 Sodium Lauryl Sulfate

Column:	Acclaim Surfactant, 5 µm
Dimension:	4.6 x 150 mm
Mobile Phase:	CH_3CN/0.1 M NH_4OAc v/v 75/25
Temperature:	25 °C
Flow Rate:	1 mL/min (0.2 mL/min to MS)
Inj. Volume:	25 µL
Detection:	MS (ESI negative)
Sample:	SLS (1 ppm)

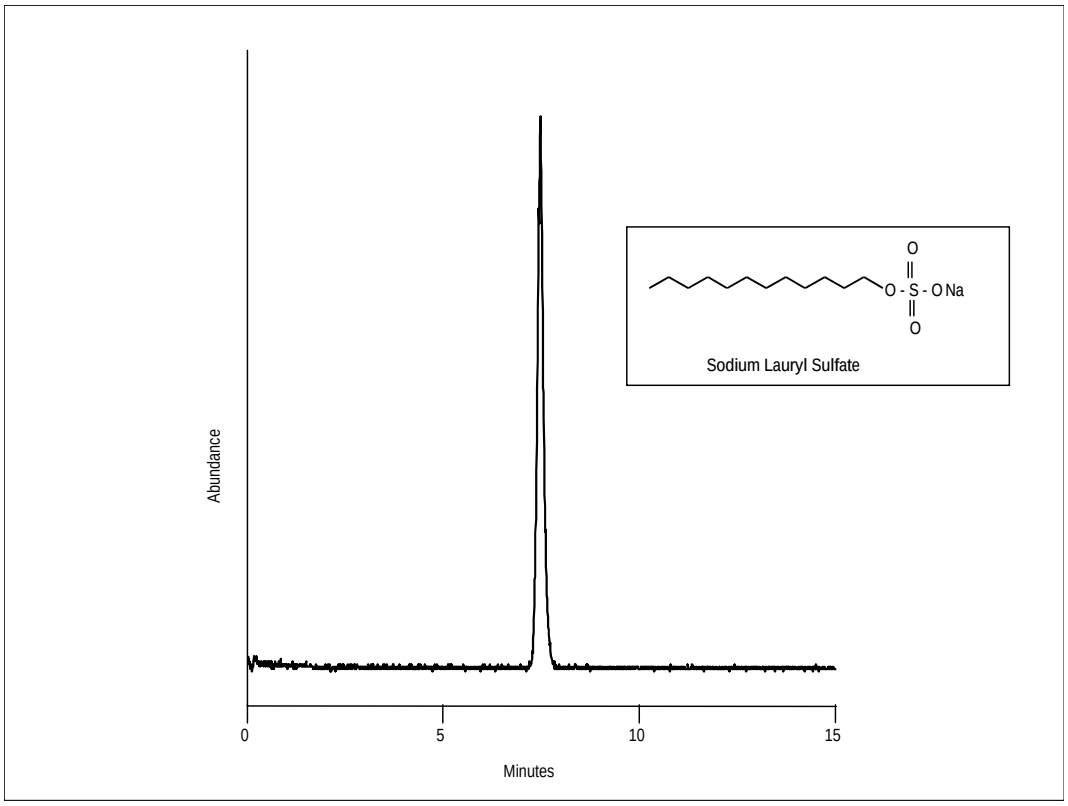


Figure 6
Analysis of Sodium Lauryl Sulfate (LC-ESI-MS)

6.1.3 POE(30) Lauryl Sulfate

Column:	Acclaim Surfactant, 5 µm
Dimension:	4.6 x 150 mm
Mobile Phase:	CH_3CN/0.5 M H_3BO_4 and 20 mM NaOH v/v 55/45
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	10 µL
Detection:	Suppressed CD (AMMS III)
Sample:	POE(30) Lauryl Sulfate (1000 ppm)

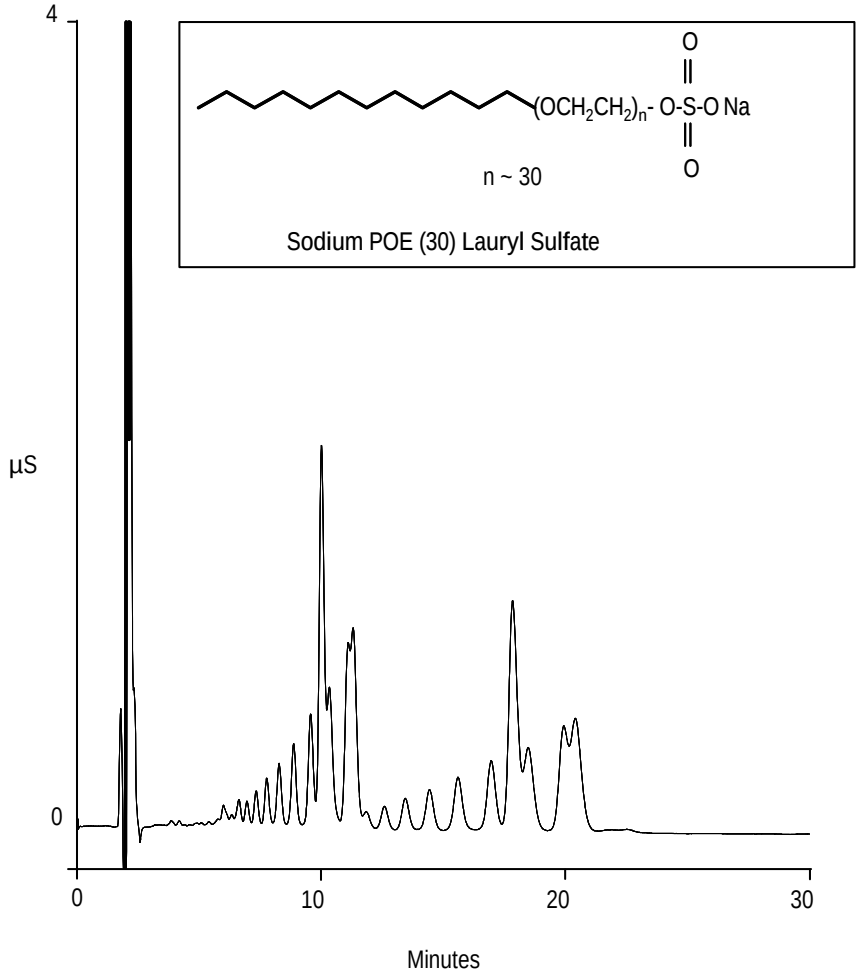


Figure 7
Analysis of POE(30) Lauryl Sulfate with Suppressed Conductivity Detection

6.2 CATIONIC SURFACTANTS

6.2.1 Cationic Surfactants with ELS Detection

Column: Acclaim Surfactant, 5 µm
Dimensions: 4.6 x 150 mm
Mobile Phase: (A) Acetonitrile
(B) 0.1 M NH_4OAc, pH 5.4
Gradient: 25% to 85% A in 30 min
Temperature: 30 °C
Flow Rate: 1 mL/min
Inj. Volume: 25 µL
Detection: ELSD

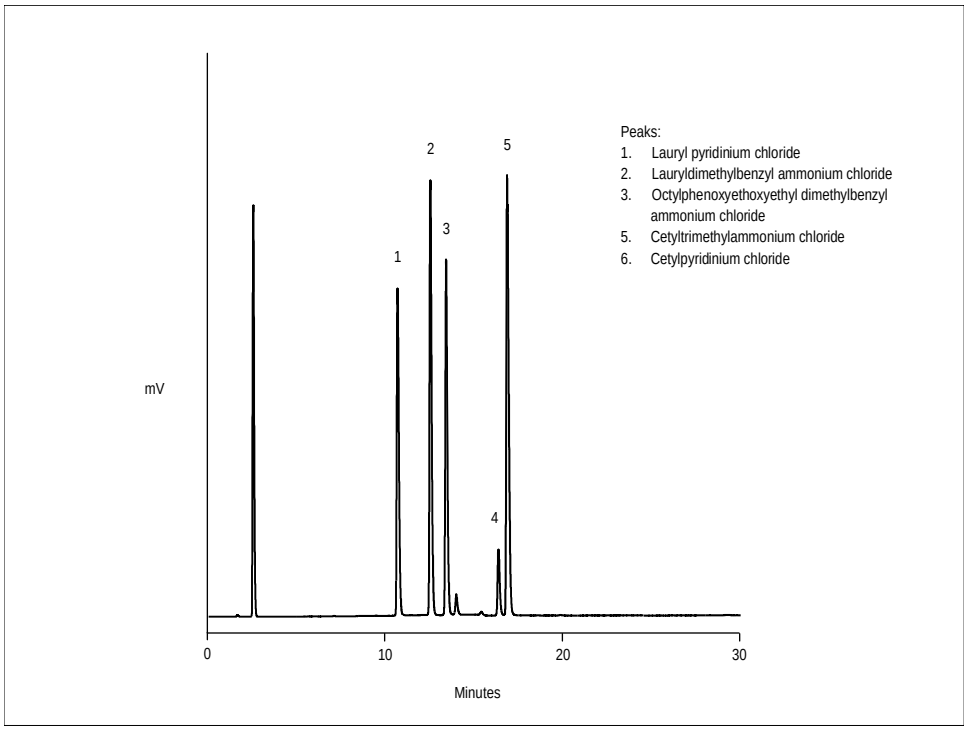


Figure 8
Analysis of Cationic Surfactants with Evaporative Light Scattering Detection

6.2.2 Cationic Surfactants with Suppressed Conductivity Detection

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	(A) Acetonitrile
	(B) 20 mM Acetic acid
Gradient:	20-50% A in 15 min
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	10 µL
Detection:	Suppressed CD (CMMS III)

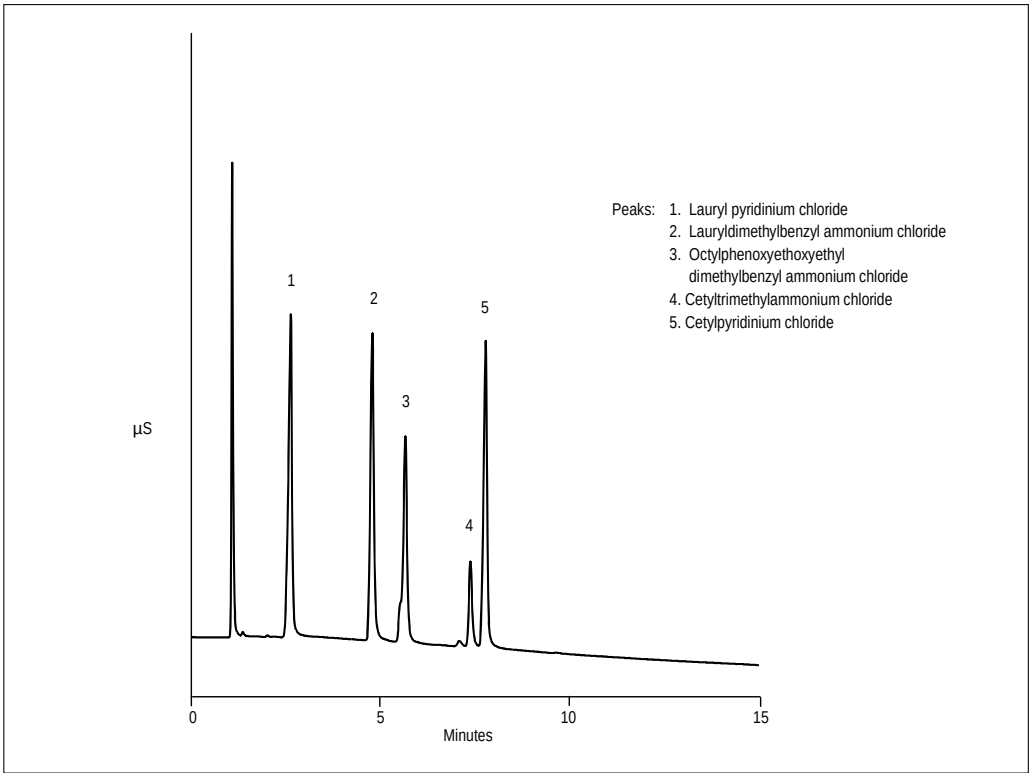


Figure 9
Separation of Cationic Surfactants with Suppressed Conductivity Detection

6.2.3 Benzalkonium Salts

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	Acetronitrile/ 0.1 M NH_4OAc, pH 5.4 v/v 60/40
Temperature:	25 °C
Flow Rate:	1 mL/min (0.2 mL/min to MS)
Inj. Volume:	5 µL
Detection:	MS (ESI positive)
Peak:	Benzyl dimethyl hydrogenated tallow ammonium chloride (10 ppm)

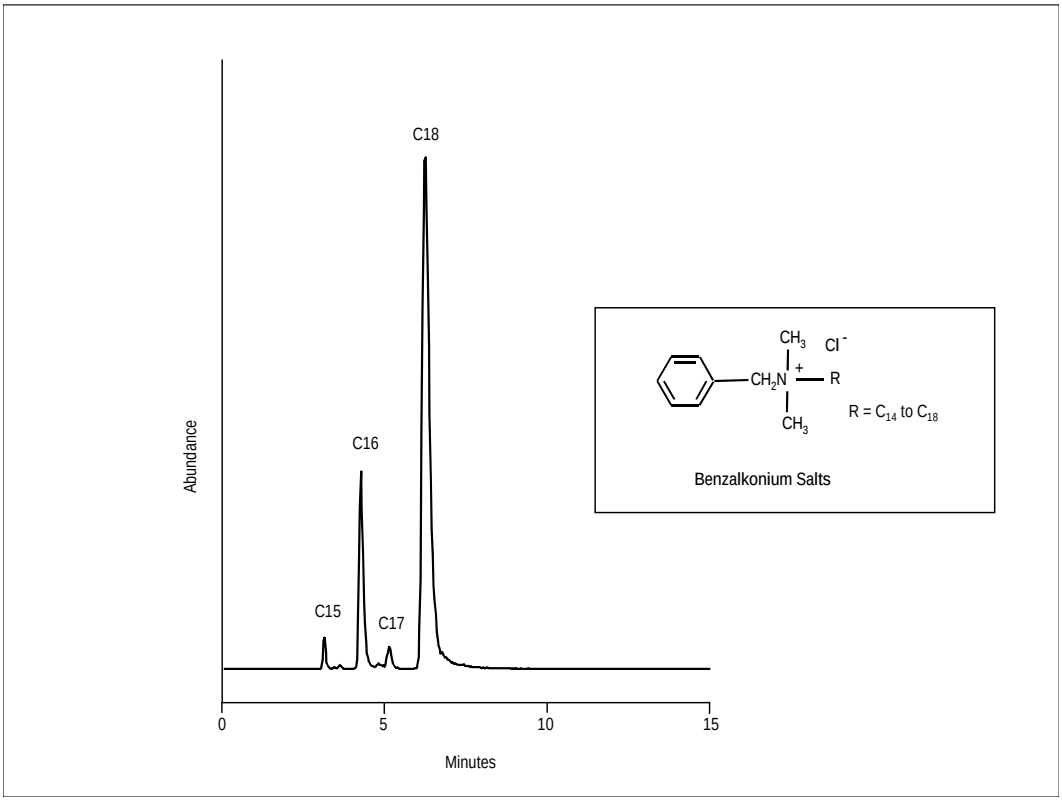


Figure 10
Analysis of Benzalkonium Salts with LC-ESI-MS Detection

6.2.4 Quaternary Imidazolium Compounds

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	(A) Acetonitrile
	(B) 0.1 M NH_4OAc, pH 5.4
Gradient:	25–85% A in 30 min
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	10 µL
Detection:	ELSD

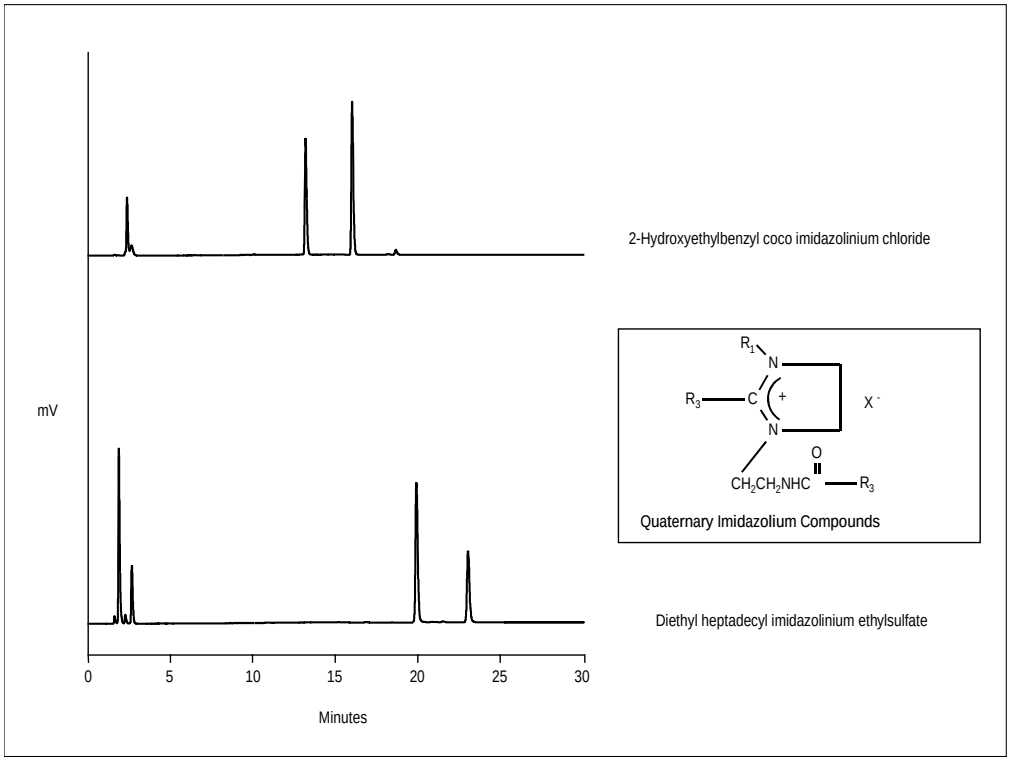


Figure 11
Analysis of Imidazolium Compounds with Evaporative Light Scattering Detection

6.3 NON-IONIC SURFACTANTS

6.3.1 Ethoxylated Quaternary Compounds

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	Acetonitrile/0.2 M NH_4OAc, pH 5.4 v/v 40/60
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	25 µL
Detection:	ELSD
Analyte:	Ethoquad 18/25 (0.2%)

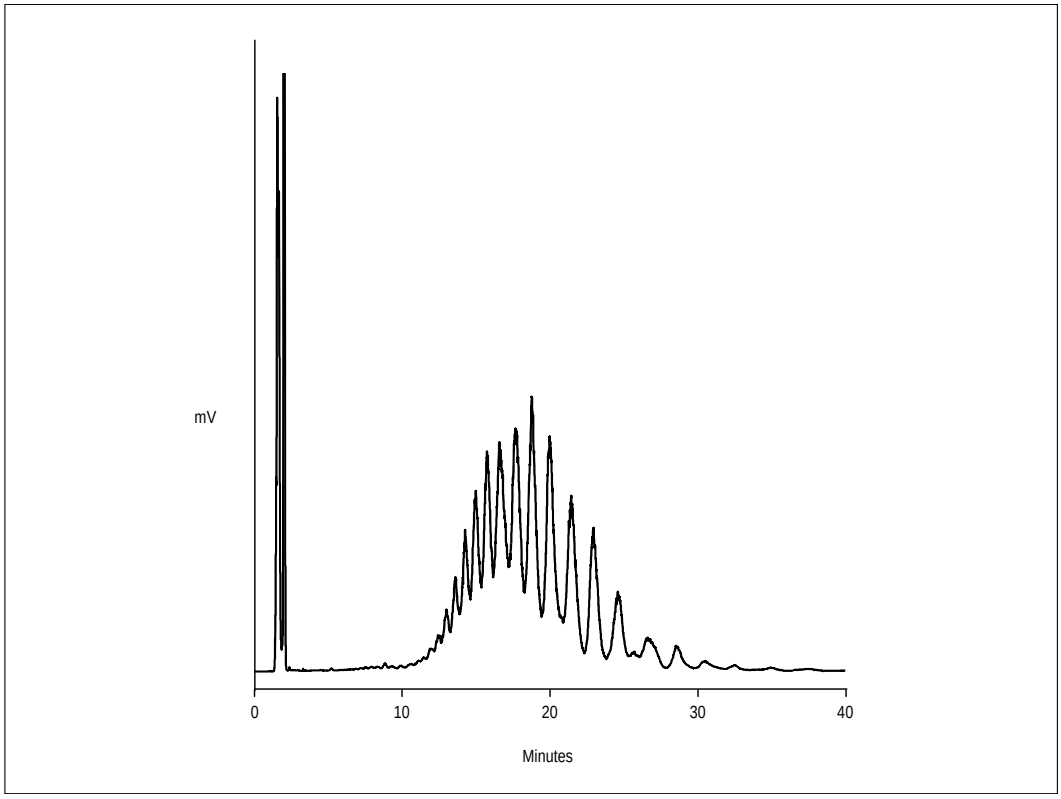


Figure 12
Analysis of Ethoxylated Quats

6.3.2 PEG Monoethyl Ether (MW-550)

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	(A) Acetonitrile
	(B) H_2O
Gradient:	5–20% A in 25 min
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj.Volume:	10 µL
Detection:	ELSD

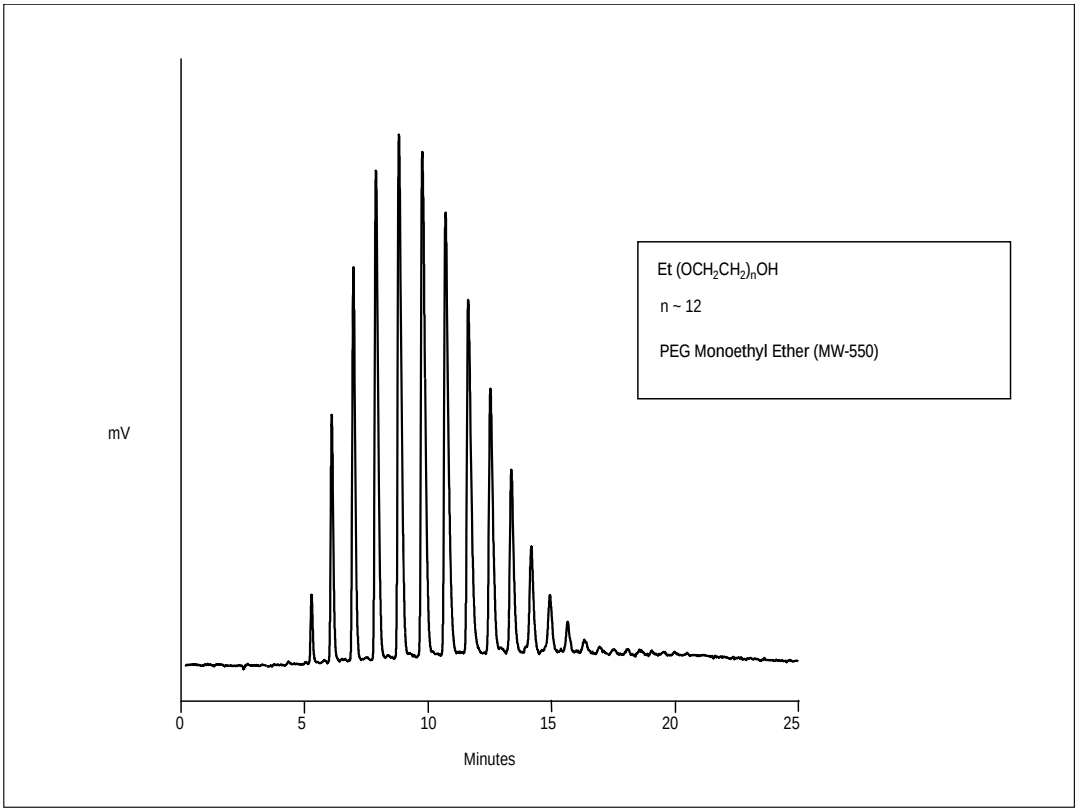


Figure 13
Analysis of PEG Monoethyl Ether (MW-550)

6.3.3 Triton X-100

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	CH_3CN/50mM NH_4OAc, pH5.4 v/v 45/55
Temperature:	25 °C
Flow Rate:	1 mL/min (0.2 mL/min to MS)
Inj. Volume:	25 µL
Detection:	MS (ESI positive)
Sample:	Triton X-100 (100 ppm)

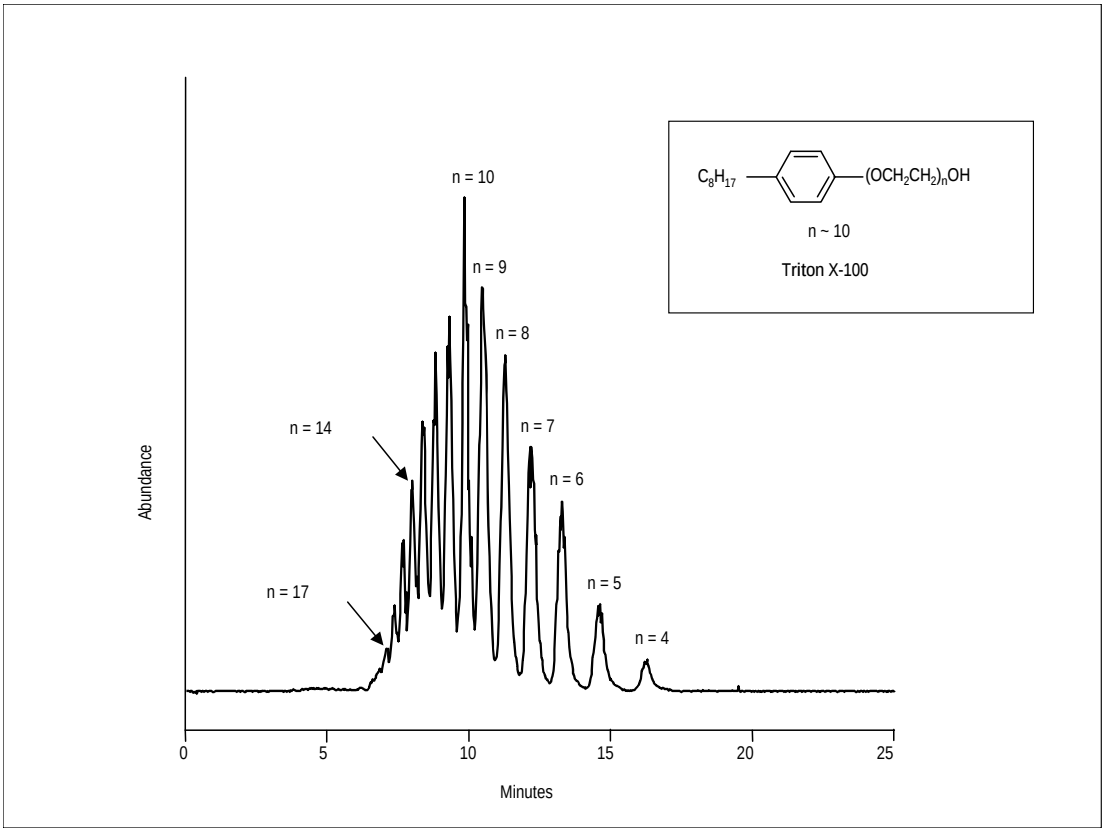


Figure 14
Analysis of Triton X-100 (LC-ESI-MS)

6.3.4 NEODOL 25-7

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	(A) Acetonitrile
	(B) H_2O
Gradient:	40–65% A in 30 min; then hold at 65%, A for 5 min
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	10 µL
Detection:	ELSD

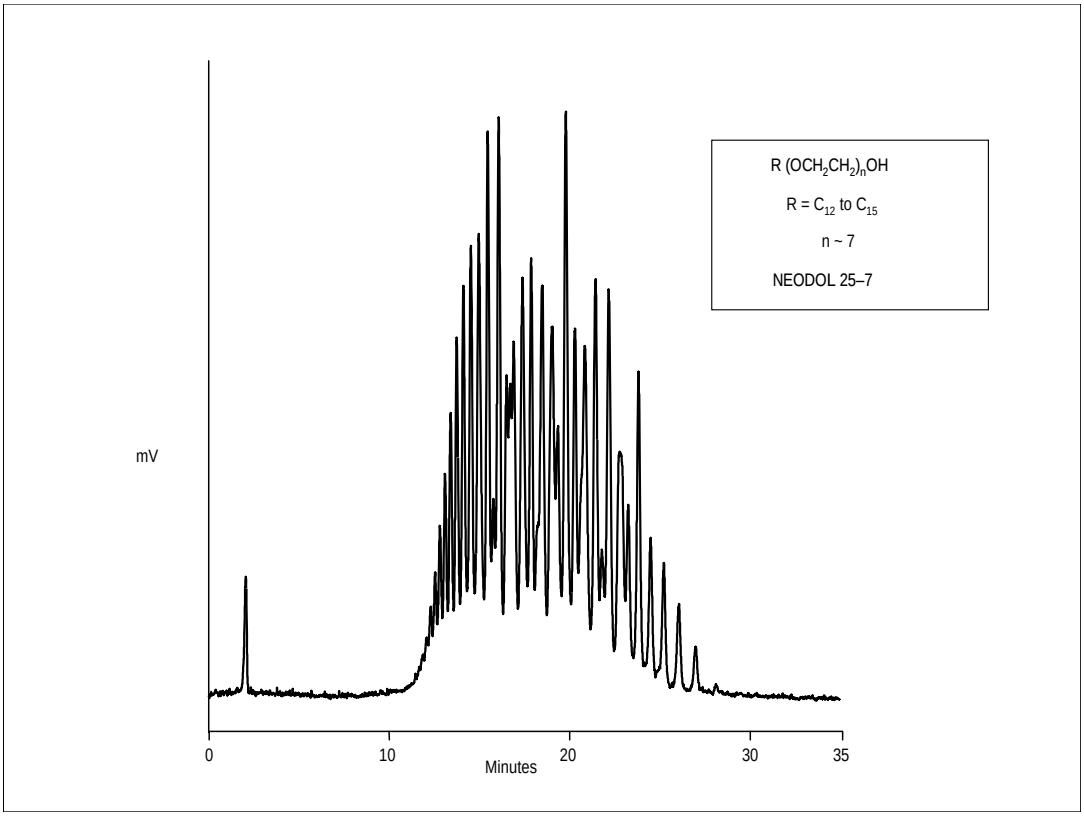


Figure 15
Analysis of Ethoxylated Alcohol (NEODOL 25-7)

6.3.5 ZONYL FSO Perfluorosurfactant

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	(A) Acetonitrile
	(B) H_2O
Gradient:	40–70% A in 30 min; then hold at 70% A for 5 min
Flow Rate:	1 mL/min
Inj. Volume:	10 µL
Detection:	ELSD

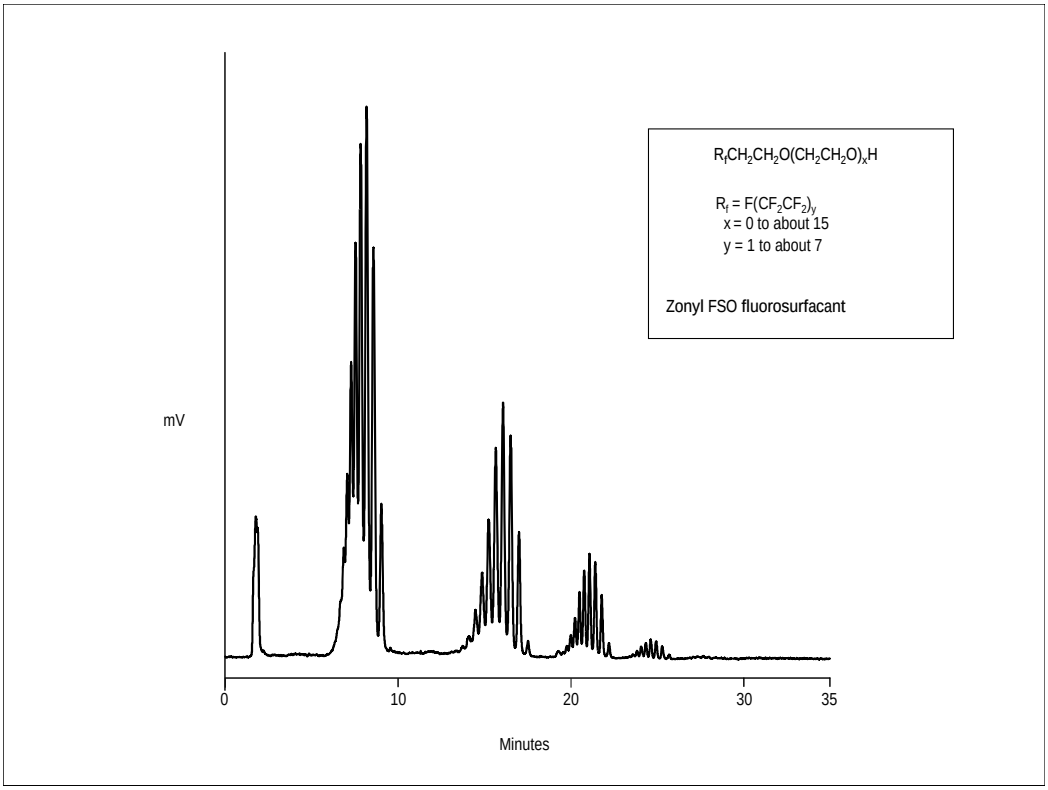


Figure 16
Analysis of ZONYL FSO Perfluorosurfactant

6.3.6 Polyethylene Glycols (PEG)

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile phase:	ACN/0.1M NH_4OAc, pH 5.4 v/v 60/40
Temperature:	30 °C
Flow rate:	1 mL/min
Injection vol.:	10 µL
Detection:	ELSD

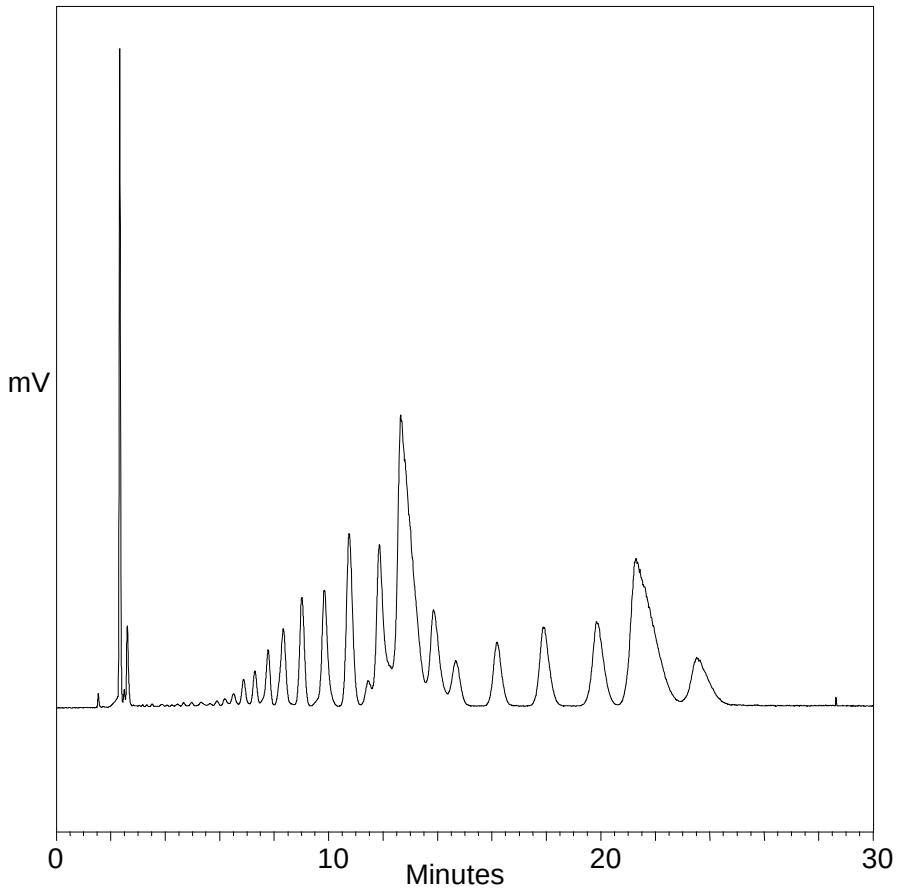


Figure 17
POE (30) Ammonium Lauryl Sulfate

6.4 SURFACTANTS IN COMMERCIAL PRODUCTS

6.4.1 Laundry Detergent

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	(A) Acetonitrile
	(B) 0.1 M NH_4OAc, pH 5.4
Gradient:	25–80% A in 30 min., then hold at 80% A for 15 min.
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	5 µL
Detection:	ELSD with UV at 225 nm
Sample Prep:	Dilute 10x with 70% acetonitrile, then filtered through 0.2-µm membrane

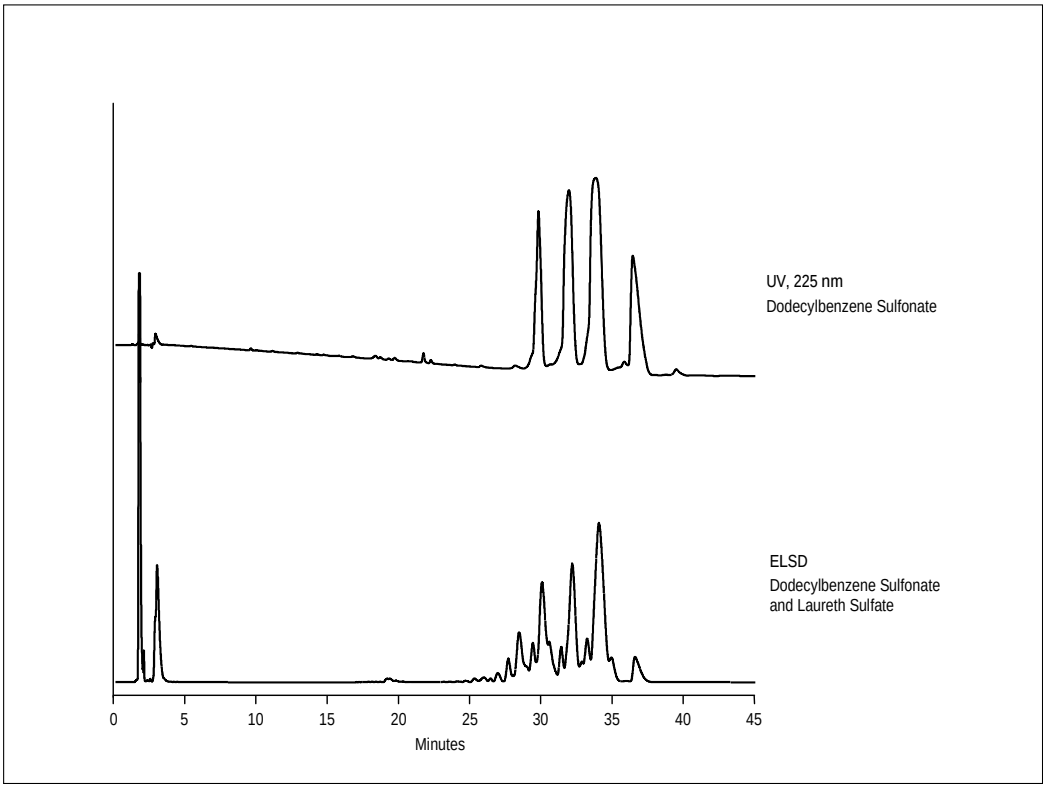


Figure 18
Analysis of a Laundry Detergent

6.4.2 Shampoo

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	(A) Acetonitrile
	(B) 0.1 M NH_4OAc, pH 5.4
Gradient:	25–80% A in 30 min; then hold at 80% A for 15 min.
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	10 µL
Detection:	ELSD and UV

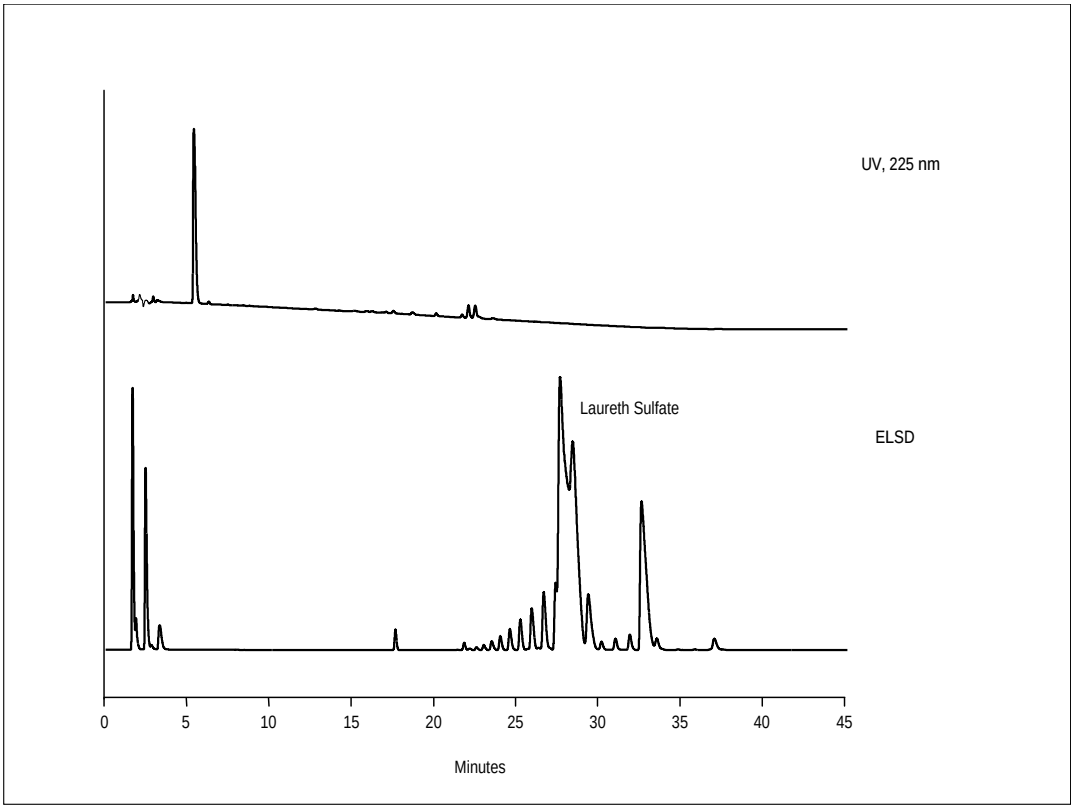


Figure 19
Analysis of Surfactants in a Shampoo

6.4.3 Fabric Softener

Column:	Acclaim Surfactant, 5 µm
Dimensions:	4.6 x 150 mm
Mobile Phase:	(A) Acetonitrile (B) THF (C) 0.1 M NH_4OAc, pH5.4
Gradient:	40% A/10% B/50% C to 75% A/10% B/75% C in 20 min
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	20 µL
Detection:	ELSD
Peaks:	*N*-methyl bis[ethyl(tallowate)]-2-hydroxyethyl ammonium methyl sulfate

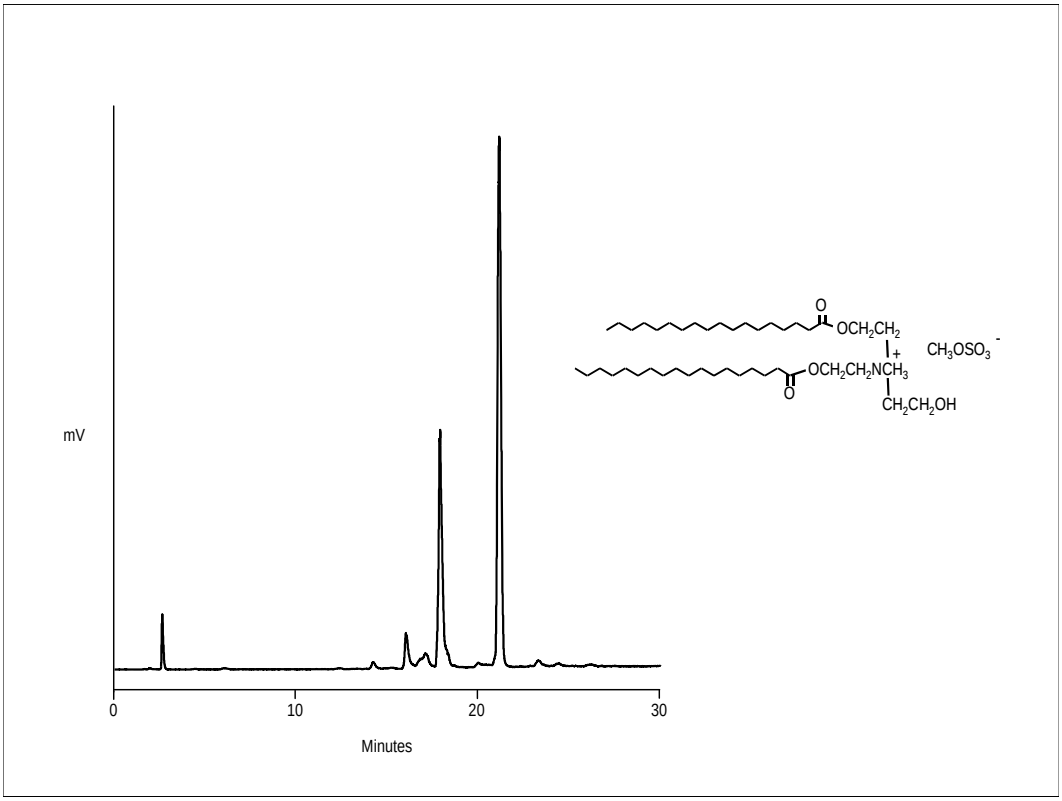


Figure 20
Analysis of Surfactants in a Fabric Softener

SECTION 7 – TROUBLESHOOTING GUIDE

The following instructions should help you to locate and eliminate problems traceable to hardware and chemistry issues. It also provides a selection of cleanup and reconditioning procedures that have been found effective by many users. Please keep in mind that some problems may be due to other reasons, such as sample contamination, poor water quality, etc. If you cannot solve your problem with the help of this manual, please contact the nearest DIONEX office (see, "DIONEX Worldwide Offices" on the Dionex Reference Library CD-ROM).

7.1 HIGH BACKPRESSURE

7.1.1 Finding the Source of High System Pressure

If the system pressure is excessively high, determine the cause of the high pressure. The system should be used with a high-pressure in-line filter for mobile phases. The filter should be positioned between the gradient pump pressure transducer and the injection valve. Make sure you have a high-pressure in-line filter in place and that it is not contaminated.

A. **Make sure that the pump is set to the correct flow rate.** Higher than recommended mobile phase 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.** It could be a piece of tubing that has plugged, collapsed tubing walls from over tightening, an injection valve with a plugged port, a column with particulates plugging the bed support, a plugged High-pressure In-line filter, or the detector cell. To identify which part of the chromatographic system is causing the problem, disconnect the pump fluid line from the injection valve and turn the pump on. Watch the pressure; it should not exceed 50 psi (0.34 MPa). Continue adding the system components back into the fluid path, one by one, while watching the system pressure.

7.2 HIGH BACKGROUND OR NOISE

7.2.1 Preparation of Mobile Phases

A. Ensure the mobile phase is made correctly.

B. Ensure the eluents are made from chemicals with the recommended purity.

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

7.2.2 A Contaminated Guard or Analytical Column

Remove the Acclaim Surfactant Analytical Column from the system. Connect the fluid lines to a piece of back pressure tubing. If the background decreases, then the column is the cause of the high background.
See Section 4.4.2, "Column Quickstart" Steps 6 through 10.

7.2.3 Contaminated Hardware

To eliminate the hardware as the source of the high background, pump deionized water with a specific resistance of 18.2 megohmcm through the system. If it is not, check the detector cell by injecting deionized water directly into it.

7.3 POOR PEAK RESOLUTION

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

7.3.1 Loss of Column Efficiency

A. Check to see if headspace has developed in the analytical column. This may be due to improper use of the column such as submitting it to high pressures. Remove the column's top end fitting (see Section 7.1.2, "Replacing Column Bed Support Assemblies"). If the resin does not fill the column body all the way to the top, the resin bed has collapsed, creating a headspace. Headspace of 1–2 mm is the maximum allowable before the column demonstrates significant losses of efficiency. If more than 2 mm of headspace is observed, the column must be replaced.

B. Extra-column system effects can result in sample band dispersion, decreasing peak efficiencies. Make sure you are using tubing with an i.d. of no greater than 0.010" to make all liquid line connections between the injection valve and the detector cell inlet on standard bore (4-mm) systems. Check for leaks.

7.3.2 Poor Resolution Due to Shortened Retention Times

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

A. Check the eluent flow rate. Determine if the actual flow rate is different than the flow rate specified by the analytical protocol. Measure the eluent flow rate after the column using a stopwatch and graduated cylinder. Allow the system to equilibrate for at least 5 minutes before making the measurement to allow time for the pump pressure feedback to engage.
B. Check to see if the mobile phase compositions and concentrations are correct. For isocratic analysis, a mobile phase buffer component that is too strong will cause the peaks to elute later. Prepare fresh buffer solution. If you are using a gradient pump to proportion the final mobile phase from concentrated solutions in two or three different reservoirs, the composition of the final mobile phase may not be accurate enough for the application. Use one reservoir containing the correct final mobile phases composition to see if proportioning accuracy is the problem. This may be a problem when one of the concentrated solutions is proportioned at less than 5%.
C. Column contamination can lead to a loss of column efficiency. Refer to Section 4.5, “Caring for the Column”, for recommended column cleanup procedures. Possible sources of column contamination are impurities in chemicals, in the deionized water, or from the sample matrix being used. Be especially careful to make sure that chemicals with recommended purity are used. The deionized water should have a specific resistance of at least 18.2 megohm-cm.

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.

7.3.3 Loss of Front End Resolution

If poor resolutions and efficiencies are observed for the very early eluting peaks near the system void volume compared to the later eluting peaks, check the following:

A. Improper mobile phase concentration. Remake the mobile phase as required for your application. Ensure that the water and chemicals used are of the required purity.
B. Column overloading. Reduce the amount of sample components injected onto the analytical column by either diluting the sample or injecting a smaller sample volume onto the column. Ensure that late eluting components, present in high concentrations, are not overloading the column.
C. Sluggish operation of the injection valve. For pneumatically driven injector valves, check the air pressure and make sure there are no gas leaks. Check for partially plugged port faces. Refer to the injector valve manual for instructions.
D. Improperly swept volumes anywhere in the system prior to the guard and analytical columns. Swap components and refit tubing connections, one at a time, in the system prior to the analytical column and test for front-end resolution after every system change.
E. Sample-related problems. Too much organic solvent or extremes of pH in the sample matrix can be a problem. Dilute the sample or adjust the pH to be similar to the mobile phase.
F. Guard cartridge. The guard cartridge should be replaced on at regular intervals. Keep a trending log of retention times, peak efficiencies, and resolution of key analytes. Replace the guard when these parameters indicate deterioration of the columns. A compacted or contaminated guard cartridge should be replaced.