thermo scientific

Table of contents



LC that takes your productivity to new heights

The collective power of chromatography

# HPLC-Charged Aerosol Detection

## Surfactants and emulsifiers applications notebook

ThermoFisher SCIENTIFIC

Table of contents



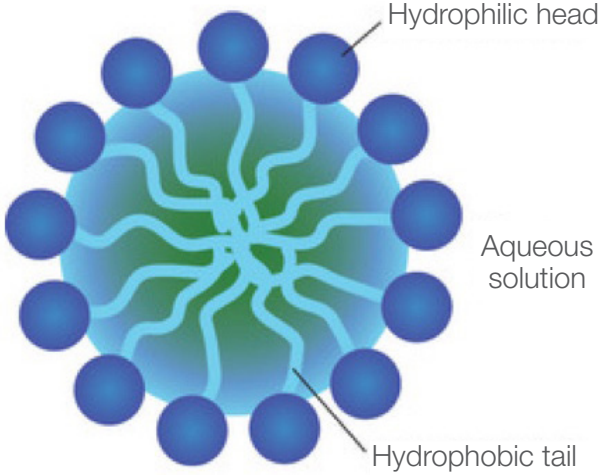
Surfactants and emulsifiers are common compounds found in everyday products, and laboratories need effective ways to develop and analyze them. This notebook offers a summary of applications and helpful information to show how liquid chromatography can be used to meet the needs of scientists working with such compounds.

## Surfactants

Surfactants (surface active agents) are compounds that contain a hydrophilic and a hydrophobic segment. When added to water or solvents, a surfactant reduces the surface tension between two immiscible solvents, a gas and a liquid, or a liquid and a solid. Surfactants have widespread uses as detergents or as cleaning, wetting, scouring, dispersing (dispersants), emulsifying, foaming, and anti-foaming agents.

## Emulsifiers

Emulsifiers are used to maintain a uniform suspension of immiscible materials. These compounds are typically surfactants and can be designed for use in specific applications and products in both the food and pharmaceutical industries.

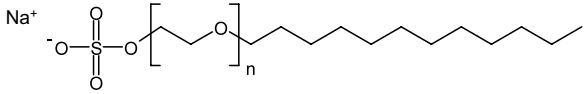


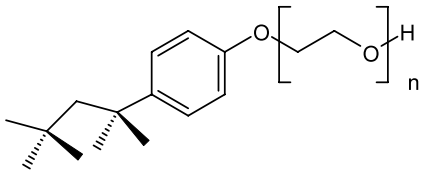

**Schematic of a micelle, which is formed by aggregation of surfactant molecules.**

# Surfactant classification



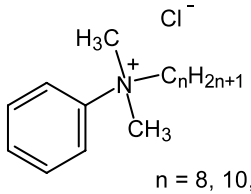
**Anionic surfactant:**
**Sodium laureth sulfate**



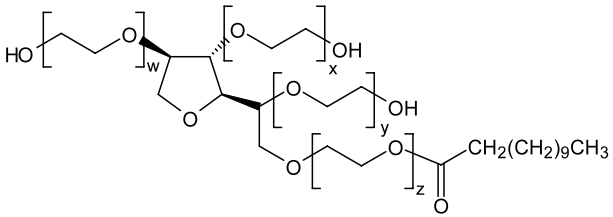




**Nonionic surfactant: Triton® X-100**

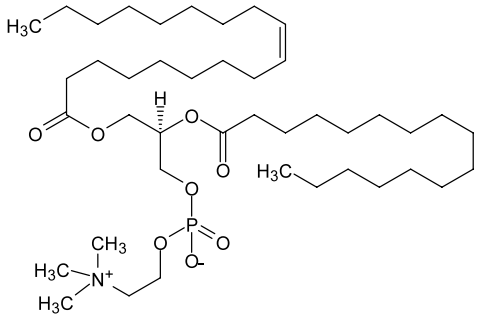
**Cationic surfactant:**
**Benzalkonium chloride**

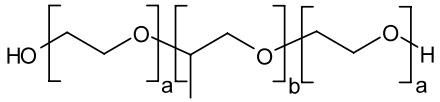



n = 8, 10, 12, 14, 16, 18




**Nonionic surfactant: Polysobate 20**




**Nonionic surfactant: Poloxamer**

**Zwitterionic surfactant: Phosphatidylcholine**

Surfactants are a diverse group of chemicals whose structures vary widely but typically contain an oil-soluble hydrophobic chain and a water-soluble hydrophilic group. Surfactants can be categorized by their structure and include both nonionic and ionic (anionic, cationic, and zwitterionic) classes.

**Nonionic surfactants** have covalently bonded oxygen-containing hydrophilic groups connected to hydrophobic parent structures and include:

- Alkylphenol ethoxylates (e.g., nonoxynols; Triton® X-100)
- Fatty acid esters of sorbitol, e.g., Spans (sorbitan monolaurate); Tweens (Tween 20, 40, 60, or 80)
- Terminally blocked ethoxylates (e.g., poloxamers)

**Anionic surfactants*** contain an anionic functional group, e.g., ammonium lauryl sulfate, sodium lauryl sulfate (also called sodium dodecyl sulfate or SDS), and sodium laureth sulfate (SLES).

**Cationic surfactants*** contain a cationic functional group, e.g., cetrimonium bromide (CTAB), cetylpyridinium chloride (CPC), and benzalkonium chloride (BAC).

**Zwitterionic (amphoteric) surfactants** have both cationic and anionic centers attached to the same molecule, e.g., phospholipids, such as phosphatidylserine, phosphatidylethanolamine, phosphatidylcholine, and sphingomyelins.

* All ionic surfactants are associated with a counter-ion that can be either monatomic (inorganic anions, such as chloride, bromide, iodide; and inorganic cations, such as alkali metal, alkaline earth metal, transition metal) or polyatomic (organic anions, such as tosyls, trifluoromethane-sulfonates, methyl sulfate; and organic cations, such as ammonium, pyridinium, triethanolamine).

thermo scientific

# Measurement

## Table of contents



Analysis of surfactants can be challenging. Not only do surfactants include a wide variety of compounds differing in their chemical structure, a specific surfactant may also be a heterogeneous mixture of numerous subspecies (e.g., Tweens) or a homologous series (often referred to as congeners).

Liquid chromatographic (LC) approaches are used to separate surfactants on the basis of ionization, chain length, chain branching, or positional isomer distribution. LC methods can be used to characterize the profile of congeners and heterogeneous mixtures. This is important, for example, when studying lot-to-lot variability, which is of particular interest in the biopharmaceutical industry.

Conversely, when trying to quantitate total amounts of surfactants, the chromatographic conditions are changed so that all the subspecies elute as a single peak. This both simplifies determination and improves limits of detection.

The preferred detection method for surfactants with chromophores is ultraviolet (UV) absorbance. However, many surfactants are weak chromophores and thus cannot be detected by UV absorbance. Mass spectrometry (MS) is often used for trace analysis and for peak identification, but is limited to only compounds that can form gas phase ions. Charged aerosol detection is universal and overcomes the limitations of both UV absorbance and MS approaches making it suitable for both exploratory work and routine analysis of surfactants.

thermoscientific

# Global surfactant market



Surfactants are used by many different industries. Selection is usually determined by special properties such as biodegradability, dermatological compatibility, effectiveness and efficiency, toxicity, or regulatory clearance for use in foods

## Industrial/Environmental

- Anti-caking and dust-control agents for fertilizers
- Herbicide and pesticide formulations
- Dispersants (for oil slicks)
- Emulsions, paints, and adhesives
- Fire extinguisher additives
- Flotation agents for purification of ore
- Fracking fluids
- Gasoline and oil additives
- Inks and de-inking agents
- Oil and gas pipeline additives
- Paint, adhesive, and sealant additives
- Road construction additives

## Personal Care/Household

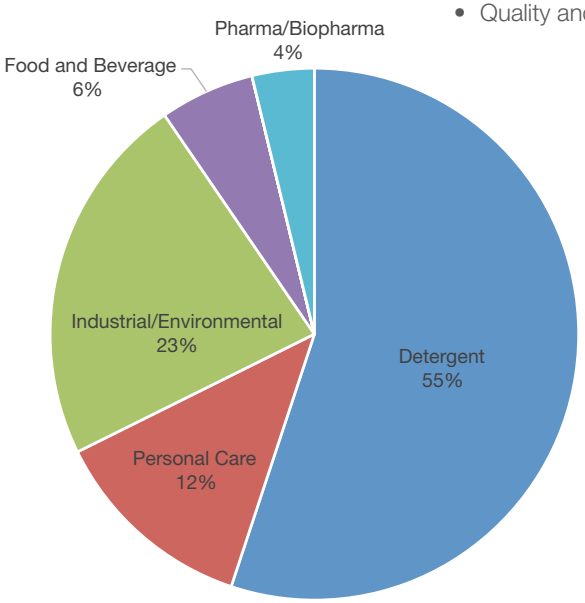
- Cosmetics
- Detergents, laundry products, fabric softeners, soaps
- Hair conditioners
- Shampoos
- Shower gels
- Soaps
- Toothpastes

## Food and Beverages

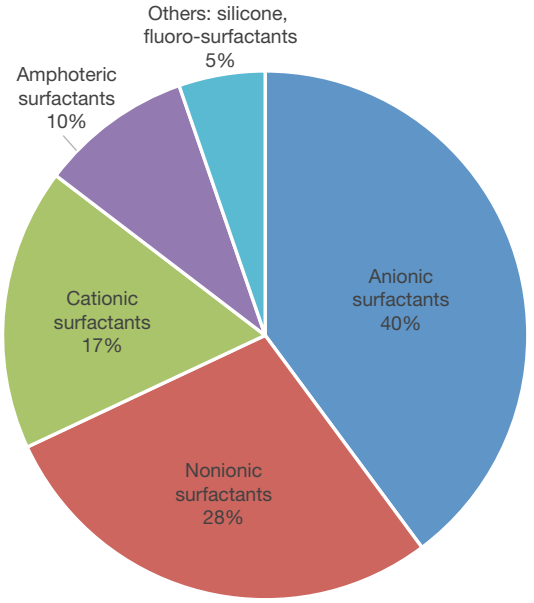
- Antimicrobials
- Defoaming agents
- Emulsifiers
- Foaming agents
- Food processing
- Mouth feel
- Quality and freshness maintenance

## Biopharma/Pharma

- Modulating solubility of active pharmaceutical ingredients (APIs)
- Influencing bioavailability of APIs
- Improving the stability of APIs in dosage forms
- Modulating immunogenic responses of APIs
- Covalent modification of peptides and proteins
- Preventing aggregation or dissociation
- Helping APIs maintain preferred polymorphic forms
- Creating emulsions including creams, ointments, liniments, pastes, films, and liquids
- Maintaining the pH and/or osmolality of liquid formulations
- Acting as antioxidants, emulsifying agents, aerosol propellants, tablet binders, and disintegrants
- Acting as the primary API of laxatives, anti-gas agents, and spermicides



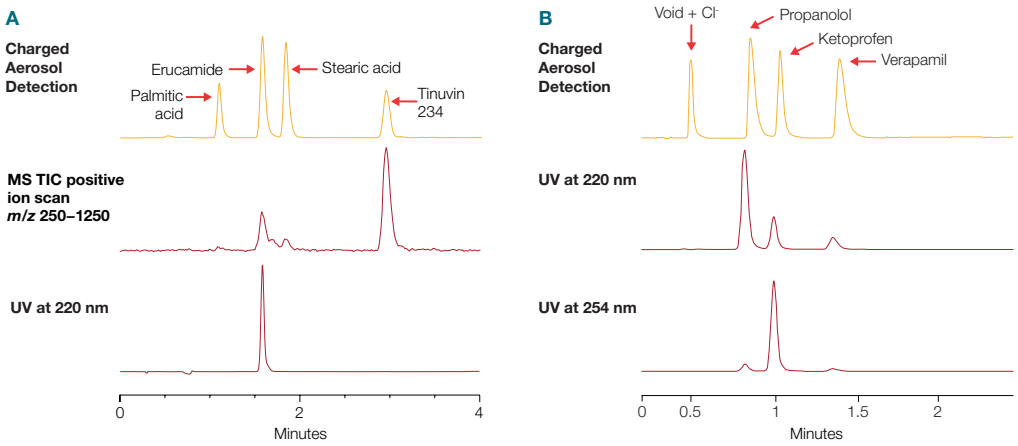

**Market distribution by application (est. for 2022)**




**Market distribution by surfactant type (est. for 2022)**

Market Report Reference: CHM081A Global Markets for Surfactant Chemicals and Materials, BCC Research, 2018

# CAD – Universal detection




**The CAD can measure all analytes in the two samples shown above.** Other detectors are more limited in scope. For example, MS requires that analytes form gas phase ions (A) while response by a UV detector depends upon the nature of the chromophore (A and B).

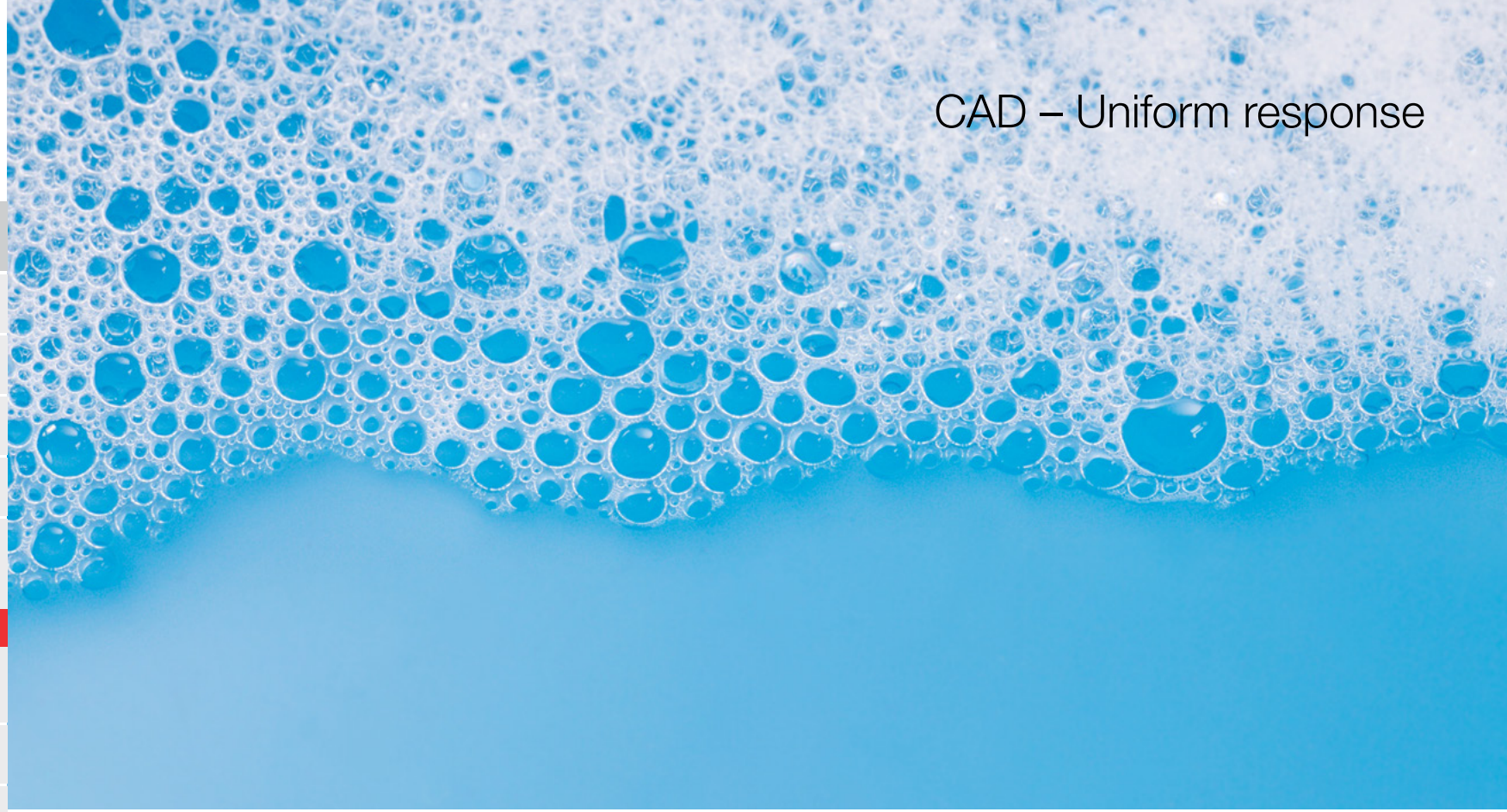
## The analyte detection challenge

No single detection method delivers ideal results for LC analysis. Often, one analyte responds more strongly to one form of detection method than another, or it may not respond at all. What is most desired is the ability to detect a wide range of analytes (universal detection) with a response that enables accurate quantitation. Charged aerosol detection is a reliable technology that will change the way you view every sample. The Charged Aerosol Detector (CAD) can detect all non-volatile, and many semi-volatile analytes, with uniform response. Charged aerosol detection has the flexibility and performance for analytical R&D, as well as the simplicity and reproducibility needed for QA/QC in manufacturing. In addition to the determination of surfactants and emulsifiers, CAD can be used for the analysis of pharmaceuticals (large and small molecule), biomolecules, food and beverages, specialty chemicals, and polymers.

Learn more: Discover what you're missing – Charged Aerosol Detector brochure

thermo scientific

# CAD – Uniform response

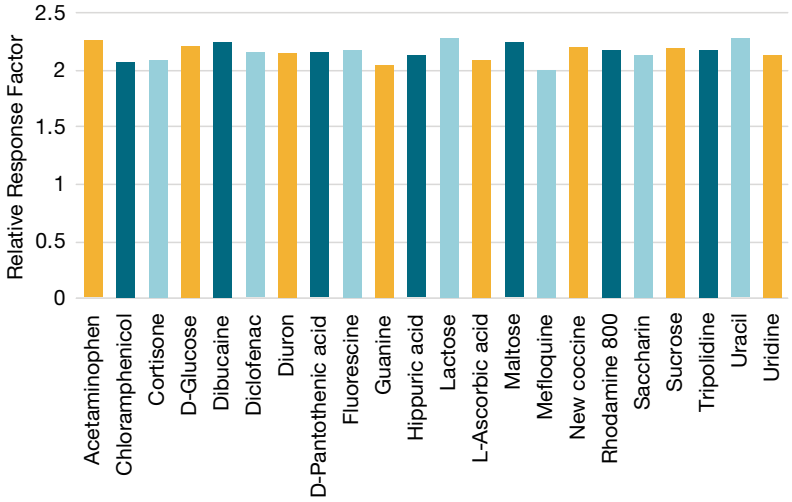


## Table of contents



## Uniform response with charged aerosol detection

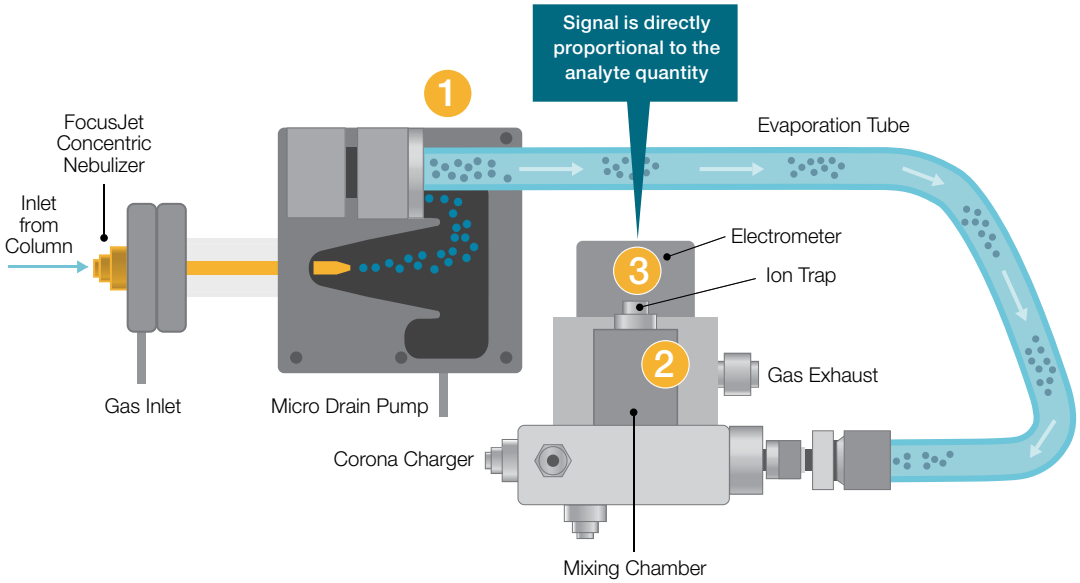
- Detector response is independent of analyte structure for all non-volatile compounds
- Excellent sensitivity coupled with wide dynamic range for unrivaled performance
- Single calibrant for quantification of multiple analytes when individual standards are not available
- Measure a surfactant as a single peak for maximum sensitivity, or profile its subspecies when determining lot-to-lot variability and for material characterization.




**The CAD shows uniform response (<5% RSD variation) among all non-volatile analytes (0.5 µg; flow injection analysis).**

# CAD – Working principles






**Schematic of CAD technology**

## Three simple steps to charged aerosol detection

### 1. Nebulization

Charged aerosol detection begins by nebulizing the column eluent into droplets, which are subsequently dried into particles. The particle size increases with the amount of analyte.

### 2. Charging

In the mixing chamber, a stream of ionized nitrogen gas collides with the analyte particles. The charge is then transferred to the particles—the larger the particle, the greater the charge.

### 3. Detection

The charged particles are transferred to a collector where the aggregate charge is measured by a highly sensitive electrometer. This generates a signal directly proportional to the mass of analyte present.

Learn more: Charged Aersol Detection Technology

thermo scientific

# CAD – Modules

## Table of contents



**Laboratories need state-of-the art instrumentation to competently analyze surfactants with charged aerosol detection.**

Thermo Scientific™ UHPLC systems combined with Thermo Scientific™ Charged Aerosol Detectors (CAD), with advanced column technologies, and with proven analytical methods provide you precise automation and advanced data handling to help you:

- Characterize many classes of surfactants and emulsifiers
- Analyze compounds in a broad range of samples
- Profile or quantify samples

**Thermo Scientific™ Vanquish™ Charged Aerosol Detectors and Thermo Scientific™ Corona™ Veo™ Charged Aerosol Detectors provide:**

- Simple, intuitive operation
- Wide linear and dynamic range
- Sub-nanogram sensitivity
- Method flexibility covering micro-flow HPLC and UHPLC applications with a single nebulizer
- Adjustable evaporation temperature to optimize signal-to-noise ratio

**Vanquish Charged Aerosol Detector**

**Corona Veo Charged Aerosol Detector**

Learn more: Discover what you're missing – Charged Aerosol Detector brochure

thermo scientific

# General purpose and specialty columns

## Table of contents



## Quality columns you can rely on for quality results

The separation and identification of surfactants and emulsifiers can be challenging due both to the diversity of surfactants and complexity of the sample matrix.

We offers quality columns with ideal selectivity in a variety of particle sizes and designs to meet all separation needs, including improved resolution, enhanced sensitivity, faster analysis, and consistent performance.

For surfactant analysis using high-sensitivity detection, the column of choice is the Thermo Scientific™ Acclaim™ Surfactant Plus for LC. Acclaim provides improved performance, versatility, and throughput for surfactant analysis with:

- Advanced surface chemistry that provides both reversed-phase and anion-exchange retention mechanisms
- Ideal selectivity for simultaneous separation of anionic, nonionic, cationic, and zwitterionic surfactants
- Excellent resolution between strongly hydrophilic compounds
- Resistance to dewetting under highly aqueous mobile phase conditions
- Exceptionally low bleed for use with charged aerosol detectors and mass spectrometers.

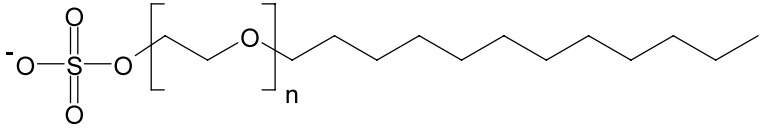

For more information: Thermo Scientific Acclaim Surfactant Plus LC Columns
For a complete selection of HPLC and UHPLC columns: HPLC and UHPLC Columns

# Anionic surfactants

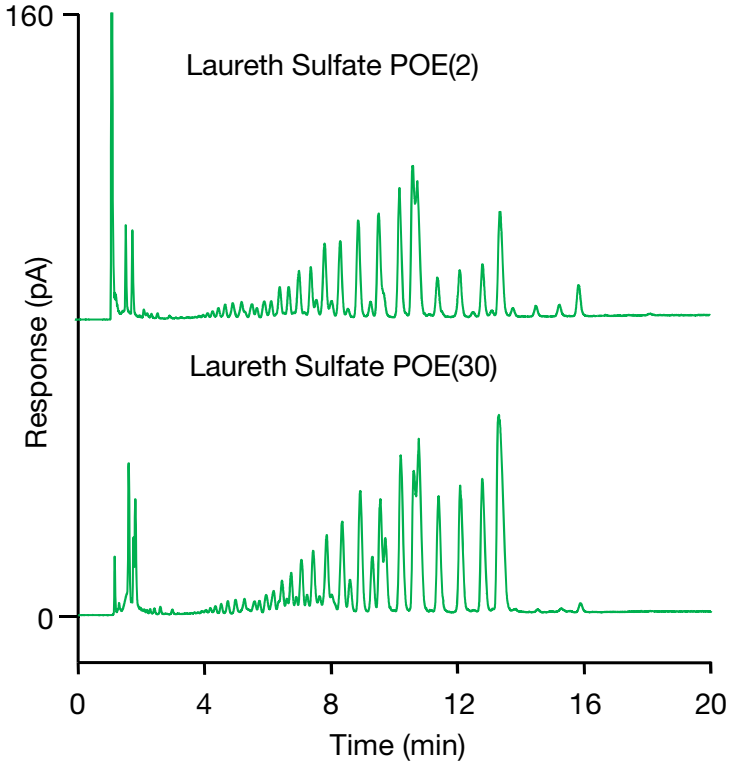


## Profiling Laureth Sulfate

| | |
|---|---|
| HPLC column: | Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm |
| Column temperature: | 30 °C |
| Mobile phase A: | Acetonitrile |
| Mobile phase B: | 100 mM Ammonium Acetate, pH 5.2 |
| Flow rate: | 0.6 mL/min |
| Gradient: | See table |
| Injection volume: | 2 µL |
| Detection: | Charged Aerosol Detector |




| Time (min) | %A | %B |
|---|---|---|
| -8 | 45 | 55 |
| 0 | 45 | 55 |
| 15 | 75 | 25 |
| 20 | 75 | 25 |




**Profiling of Laureth Sulfate POE(2) and POE(30) (10 mg/mL each)**

For more information: HPLC-CAD method for laureth sulfates

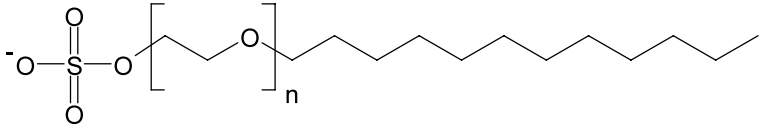
# Anionic surfactants

## Table of contents

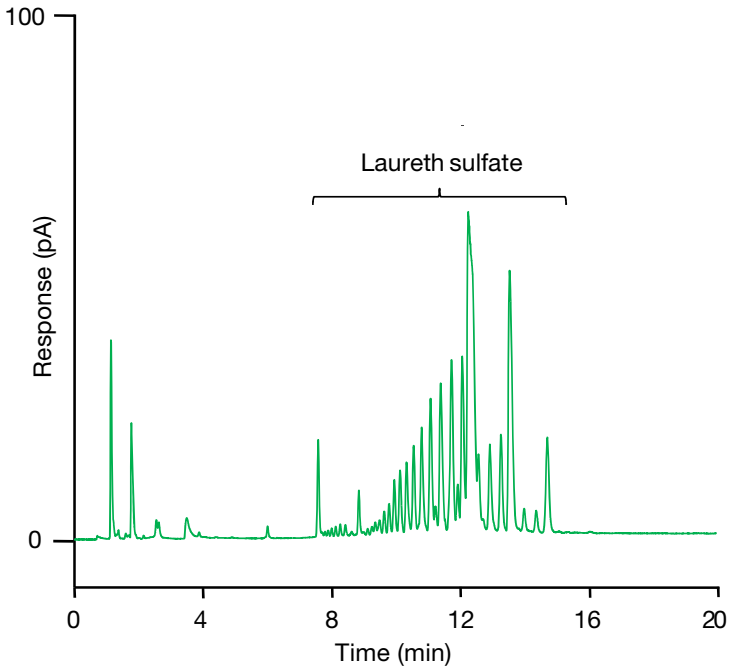


## Sodium Laureth Sulfate (SLES) in shampoo

| | |
|---|---|
| HPLC column: | Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm |
| Column temperature: | 30 °C |
| Mobile phase A: | Acetonitrile |
| Mobile phase B: | 50 mM Ammonium Acetate, pH 5.2 |
| Flow rate: | 0.6 mL/min |
| Gradient: | See table |
| Injection volume: | 2 µL |
| Detection: | Charged Aerosol Detector |




| Time (min) | %A | %B |
|---|---|---|
| -8 | 25 | 75 |
| 0 | 25 | 75 |
| 10 | 80 | 20 |
| 20 | 80 | 20 |




**Analysis of shampoo sample (40x dilution and filtered)**

For more information: HPLC-CAD for analysis of lauryl ether sulfates in shampoo

thermo scientific

# Anionic surfactants

## Table of contents

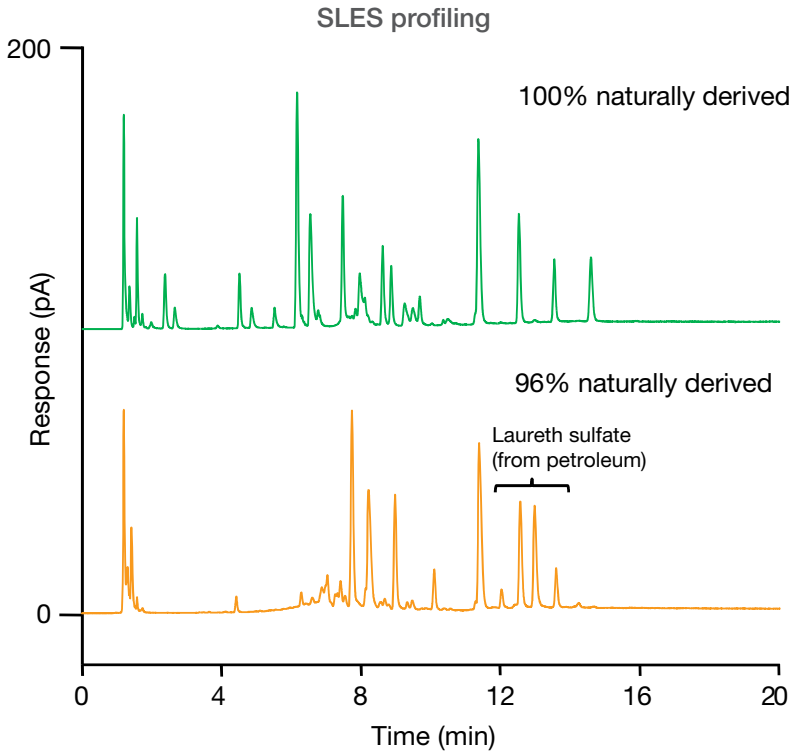


## SLES in laundry detergent

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm
Column temperature: 30 °C
Mobile phase A: Acetonitrile
Mobile phase B: 100 mM Ammonium Acetate, pH 5.2
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 2 µL
Detection: Charged Aerosol Detector

**Method development**

| Time (min) | %A | %B |
|---|---|---|
| -8 | 25 | 75 |
| 0 | 25 | 75 |
| 10 | 80 | 20 |
| 20 | 80 | 20 |




**Profiling surfactants in laundry detergents**

For more information: Analysis of two laundry detergents

# Anionic surfactants

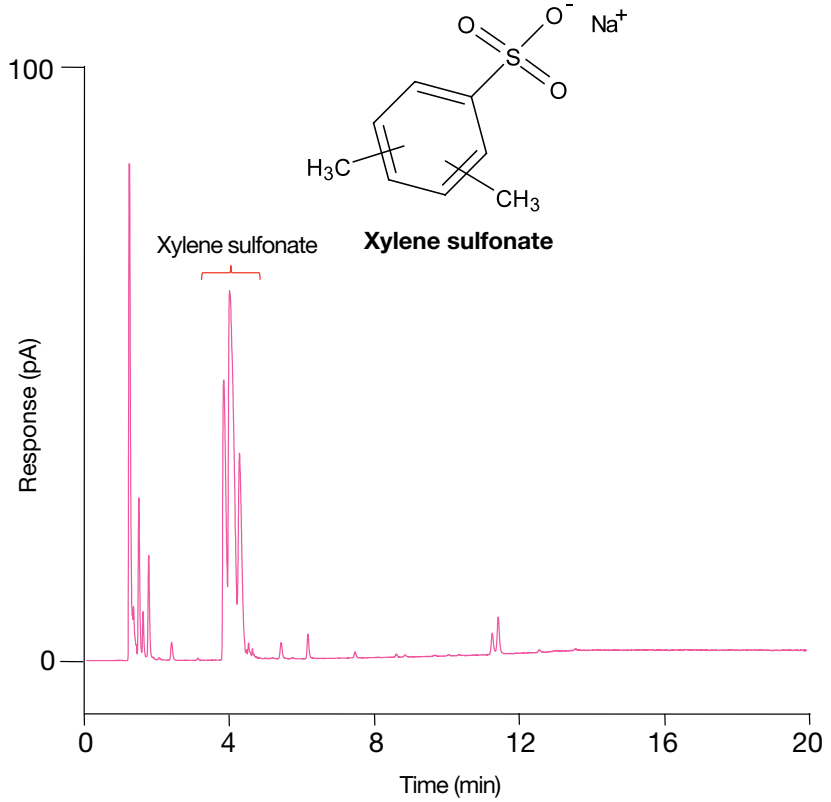


## Xylene sulfonate in foaming liquid hand soap

| | |
|---|---|
| HPLC column: | Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm |
| Column temperature: | 30 °C |
| Mobile phase A: | Acetonitrile |
| Mobile phase B: | 100 mM Ammonium Acetate, pH 5.2 |
| Flow rate: | 0.6 mL/min |
| Gradient: | See table |
| Injection volume: | 1 µL |
| Detection: | Charged Aerosol Detector |

| Time (min) | %A | %B |
|---|---|---|
| -8 | 25 | 75 |
| 0 | 25 | 75 |
| 10 | 80 | 20 |
| 20 | 80 | 20 |

While various surfactants are used in liquid hand soaps, xylene sulfonate, a highly hydrophilic hydrotrope, is the main component in a high-foaming liquid hand soap.




**Xylene sulfonate**

**Profiling surfactants in foaming hand soap**

For more information: Analysis of liquid hand soap

thermoscientific

# Cationic surfactants

## Table of contents

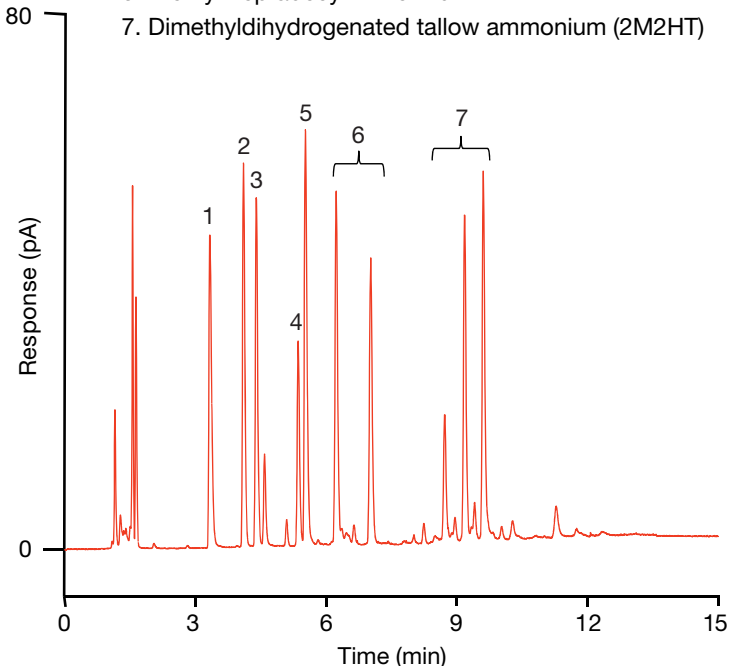


## Profiling different cationic surfactants

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm
Column temperature: 30 °C
Mobile phase A: 100 mM Ammonium Acetate, pH 5.2
Mobile phase B: Acetonitrile
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 5 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| -8 | 65 | 35 |
| 0 | 65 | 35 |
| 8 | 15 | 85 |
| 15 | 15 | 85 |

Peaks: (200–400 µg/mL each)
1. Lauryl pyridinium
2. Lauryldimethylbenzyl ammonium
3. Octylphenoxyethoxyethyl-dimethylbenzyl ammonium
4. Cetyltrimethylammonium
5. Cetylpyridinium
6. Diethyl heptadecyl imizolinium
7. Dimethyldihydrogenated tallow ammonium (2M2HT)




**High selectivity screening of different cationic surfactants**

For more information: Analysis of cationic surfactants

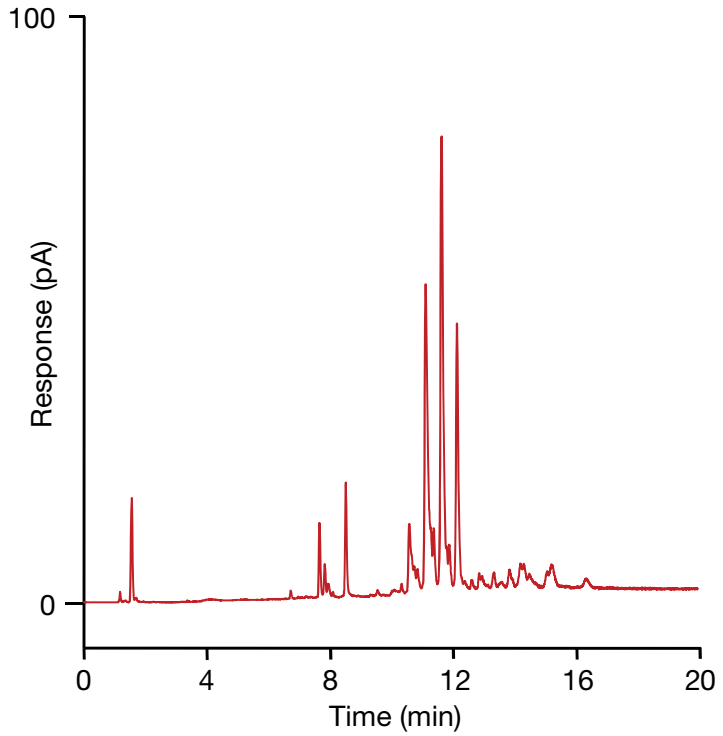
# Cationic surfactants



## Fabric softener

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm
Column temperature: 30 °C
Mobile phase A: 100 mM Ammonium Acetate, pH 5.0
Mobile phase B: Acetonitrile
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 1 µL
Detection: Charged Aerosol Detector

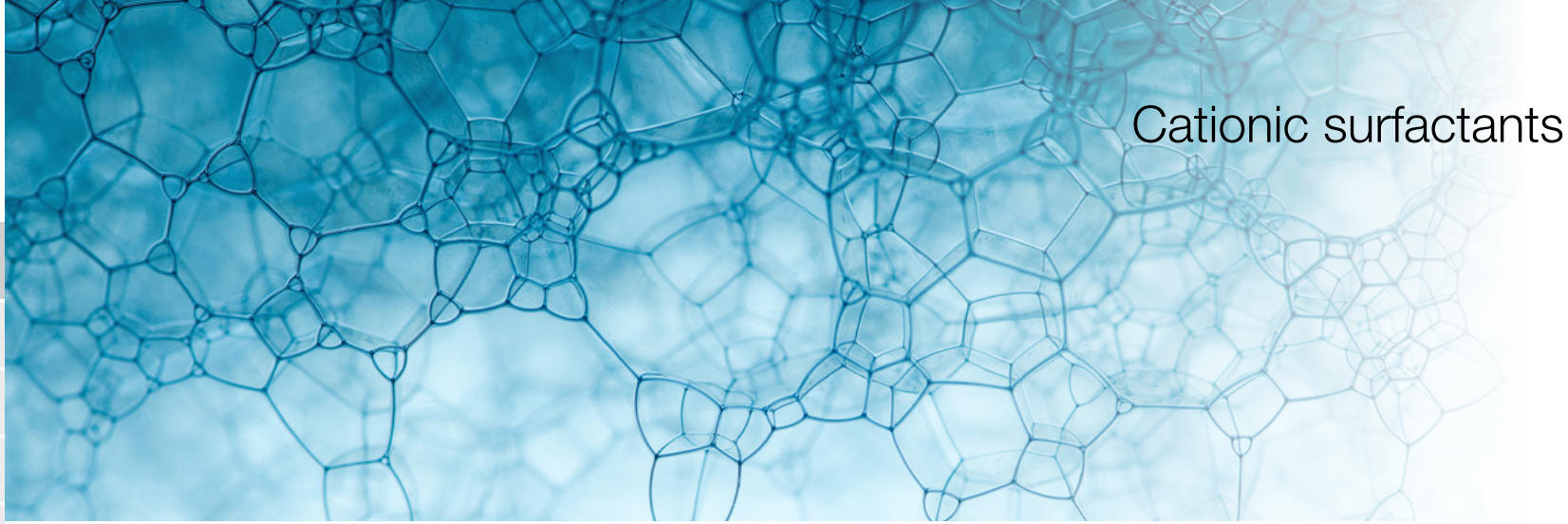
| Time (min) | %A | %B |
|---|---|---|
| -10 | 70 | 30 |
| 0 | 70 | 30 |
| 10 | 15 | 85 |
| 20 | 15 | 85 |




**Profiling cationic surfactants in fabric softener**

For more information: Analysis of fabric softner

thermoscientific



# Cationic surfactants

## Table of contents

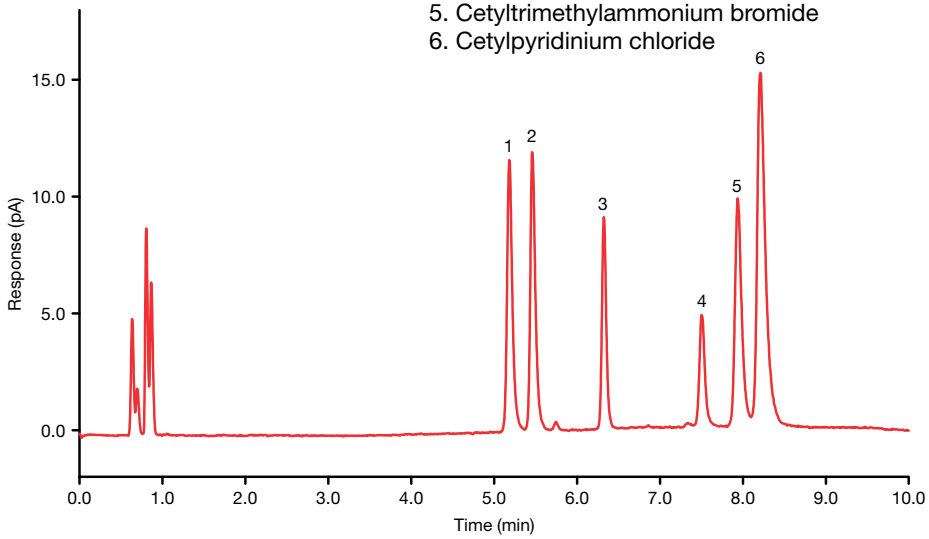


## Profiling different quaternary amines

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 2.1 × 150 mm
Column temperature: 30 °C
Mobile phase A: 100 mM Ammonium Acetate, pH 5.2/Isopropanol (95:5 v/v)
Mobile phase B: Acetonitrile/Isopropanol (95:5 v/v)
Flow rate: 0.4 mL/min
Gradient: See table
Injection volume: 2 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| 0 | 80 | 20 |
| 0.5 | 80 | 20 |
| 4 | 60 | 40 |
| 7 | 80 | 20 |
| 10 | 80 | 20 |

Peaks:
1. Dodecyltrimethylammonium bromide
2. Dodecylpyridinium chloride
3. Banzalkonium chloride (n = $C_{12}$)
4. Benzalkonium chloride (n = $C_{14}$)
5. Cetyltrimethylammonium bromide
6. Cetylpyridinium chloride




**Highly selective separation of a range of quaternary amines**

Download Application Note: Analysis of Cationic Surfactants on the Acclaim Surfactant Plus HPLC Column

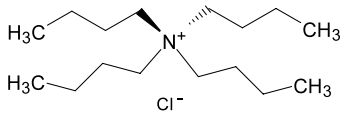
# Cationic surfactants



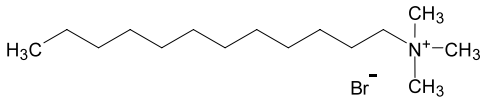
## Quaternary amines – rapid analysis

| | |
|---|---|
| HPLC column: | Accucore Vanquish C18 column, 1.5 µm, 2.1 × 100 mm (P/N 17101-102130) |
| Column temperature: | 40 °C |
| Mobile phase A: | 0.1% aq Glacial Acetic Acid |
| Mobile phase B: | Acetonitrile |
| Flow rate: | See table |
| Gradient: | See table |
| Injection volume: | 2 µL |
| Detection: | Charged Aerosol Detector |

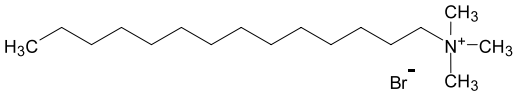
| Time (min) | Flow rate (mL/min) | %A | %B |
|---|---|---|---|
| -1.5 | 0.50 | 90 | 10 |
| 0 | 0.60 | 90 | 10 |
| 0.2 | 0.50 | 30 | 70 |
| 0.8 | 0.47 | 25 | 75 |
| 0.9 | 0.45 | 0 | 100 |
| 1.0 | 0.45 | 0 | 100 |
| 1.2 | 0.5 | 90 | 10 |
| 2.0 | 0.5 | 90 | 10 |



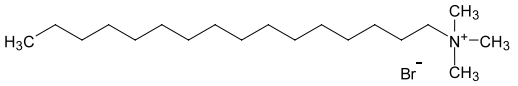

Tetrabutylammonium chloride



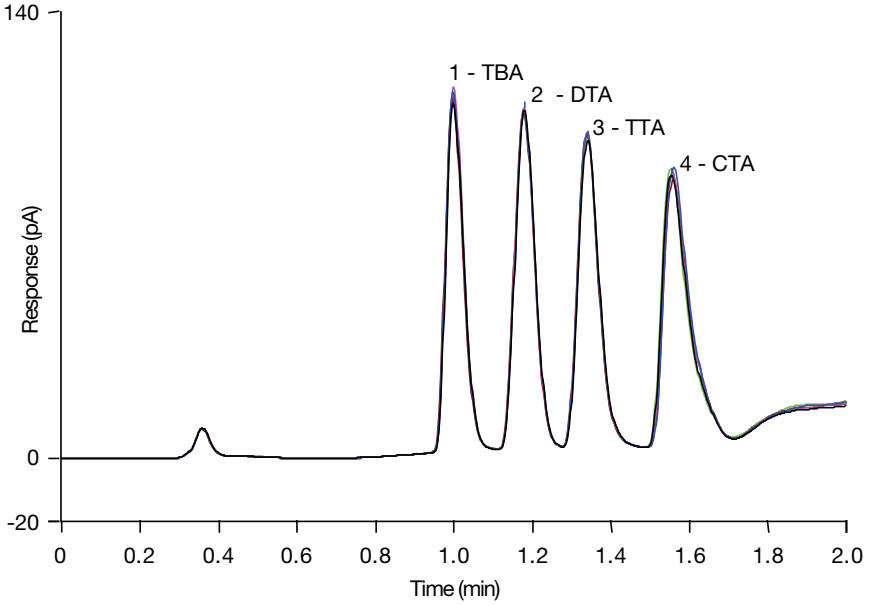

Dodecyltrimethylammonium (DTA) bromide




Tetradecyltrimethylammonium (TTA) bromide




Hexadecyltrimethylammonium (CTA) bromide




**Rapid analysis of four cationic surfactants (n=5)**

Download Poster: Fast and Sensitive Determination of Quaternary Amines by UHPLC

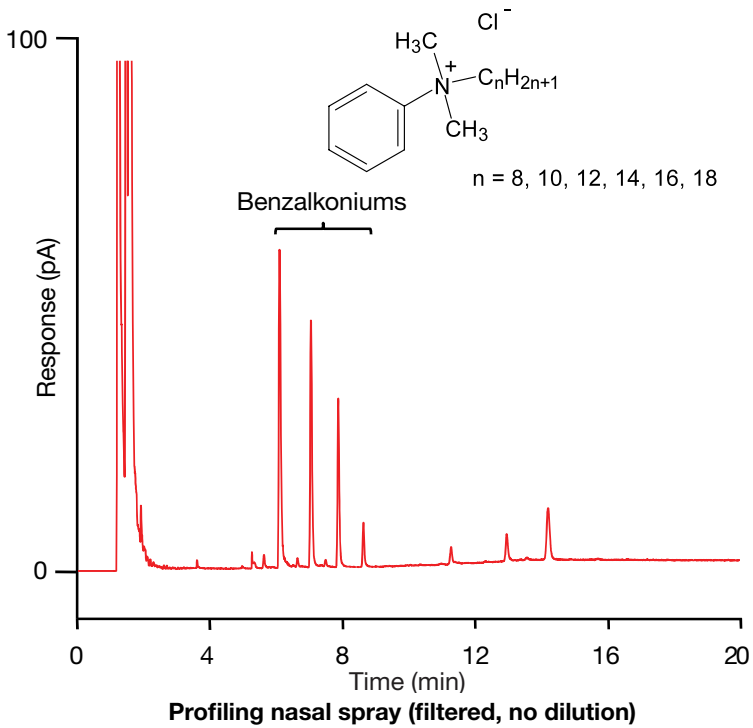
# Cationic surfactants



## Quaternary amines in nasal spray

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm
Column temperature: 30 °C
Mobile phase A: 100 mM Ammonium Acetate, pH 5.2
Mobile phase B: Acetonitrile
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 5 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| -10 | 75 | 25 |
| 0 | 75 | 25 |
| 10 | 20 | 80 |
| 20 | 20 | 80 |




**Profiling nasal spray (filtered, no dilution)**

For more information: Analysis of benzalkonium in nasal spray

thermoscientific

# Cationic surfactants

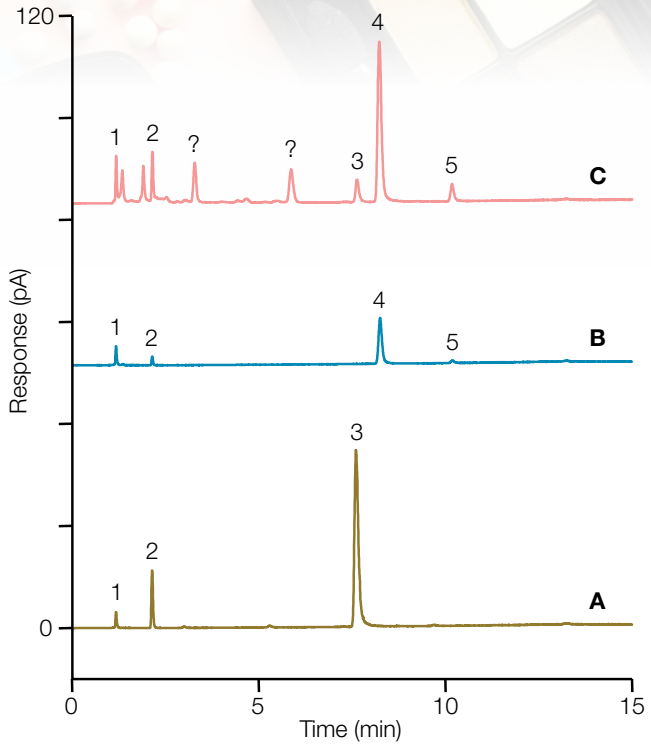
## Table of contents



## Quaternary amines in cosmetics

| | |
|---|---|
| HPLC column: | Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm |
| Temperature: | 30 °C |
| Mobile phase A: | Methanol |
| Mobile phase B: | Water |
| Mobile phase C: | 6.35 g/L Ammonium Formate, 6.00 g/L Formic Acid; pH 3.5 |
| Flow rate: | 0.6 mL/min |
| Gradient: | See table |
| Injection volume: | 2 µL |
| Detection: | Charged Aerosol Detector |

| Time (min) | %A | %B | %C |
|---|---|---|---|
| -8 | 50 | 25 | 25 |
| 0 | 50 | 25 | 25 |
| 10 | 50 | 25 | 25 |
| 15 | 75 | 0 | 25 |
| 15 | 95 | 0 | 5 |
| 17 | 95 | 0 | 5 |




**UHPLC-CAD analysis of: A) Arquad 16-29, 0.1% dilution in water; B) Variquat® PATC, 100 µg/mL in water; C) Cosmetic Formulation, 10% dilution in water. 1. Sodium; 2. Chloride; 3. Hexadecyl trimethylammonium; 4. Hexadecylamidopropyl trimethylammonium; 5. Octadecylamidopropyl trimethylammonium**

# Cationic surfactants

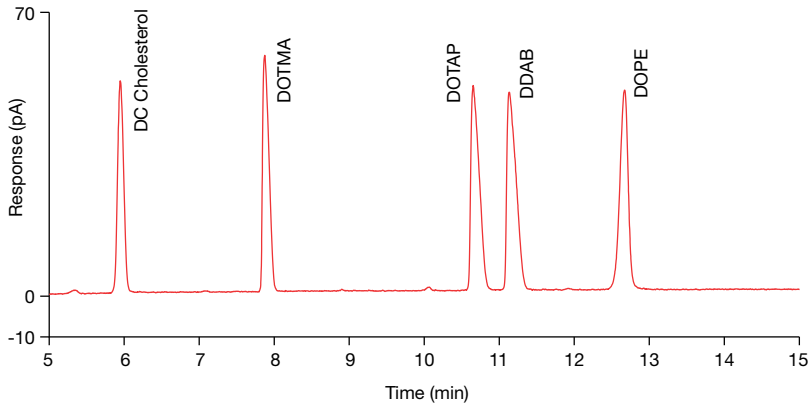


## Measurement of cationic lipid purity used as transfection agents for siRNA

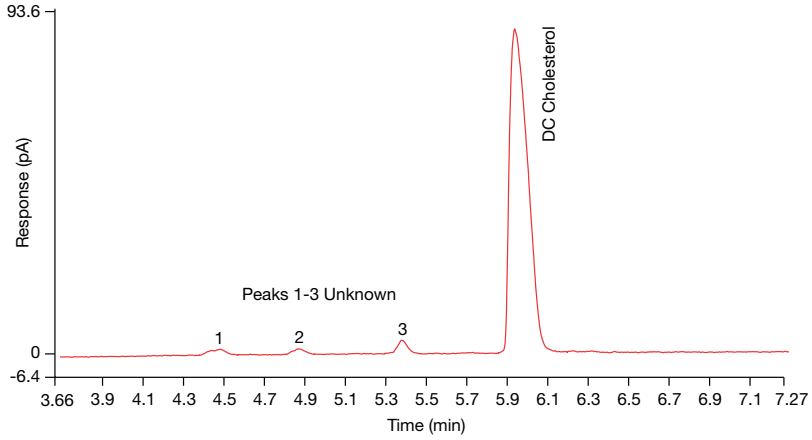
This HPLC-CAD method enables purity assessment and quantitation of several commonly used cationic lipids including, 3β-[N-(N',N'-dimethylaminoethane)-carbamoyl] cholesterol hydrochloride (DC-Chol), 1,2-dioleoyl-3-trimethylammonium-propane (chloride salt) (DOTAP), dimethyldioctadecylammonium bromide (DDAB), 1,2-di-O-octadecenyl-3-trimethylammonium propane (chloride salt) (DOTMA), and 1,2-dioleoyl-sn-glycero-3-phosphoethanolamine (DOPE).

| | |
|---|---|
| HPLC column: | C8, 2.7 µm, 4.6 × 150 mm |
| Column temperature: | 45 °C |
| Mobile phase A: | DI Water/Methanol/Trifluoroacetic Acid (600:400:1) |
| Mobile phase B: | Ethanol/Tetrahydrofuran/Trifluoroacetic Acid (750:250:1) |
| Flow rate: | 0.5 mL/min |
| Gradient: | See table |
| Injection volume: | 2 µL |
| Detection: | Charged Aerosol Detector |

| Time (min) | %A | %B |
|---|---|---|
| 0 | 55 | 45 |
| 20.0 | 30 | 70 |
| 20.1 | 55 | 45 |
| 25.0 | 55 | 45 |




**Cationic lipids standards (2.5 µg on column)**




**DC-cholesterol standard was found to be 96.7% pure**

For more information: Purity of the liposomal biotherapeutic delivery compound

thermo scientific

# Zwitterionic surfactants

## Table of contents



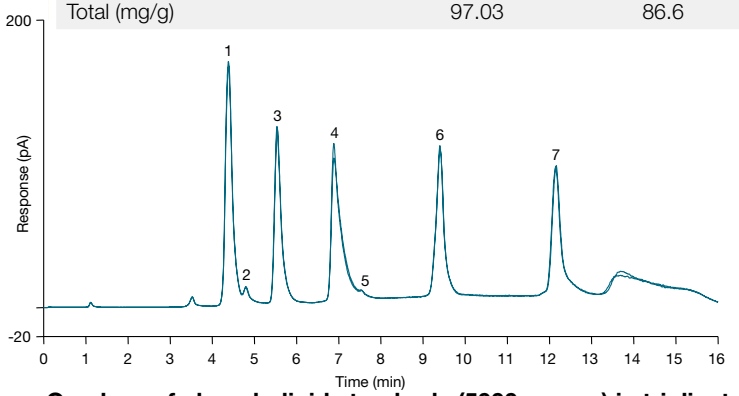
## Lecithin (egg)

General method for the measurement of different phospholipids.

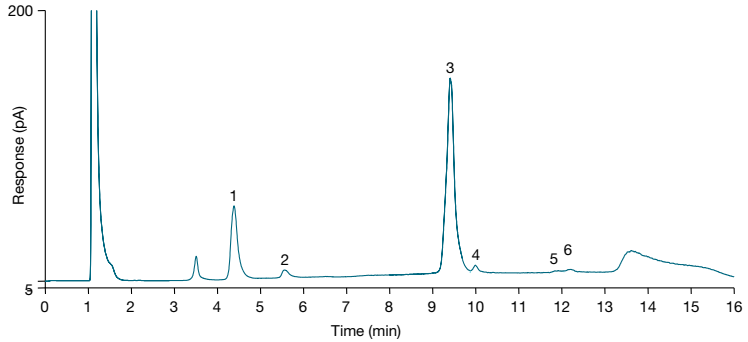
HPLC column: Thermo Scientific™ Hypersil™ silica, 5 µm, 3.0 × 150 mm
Column temperature: 50 °C
Mobile phase A: 0.5% Diethylamine Formate pH 3.0
Mobile phase B: Isopropanol
Mobile phase C: iso-Octane
Flow rate: See table
Gradient: See table
Injection volume: 2–10 µL
Detection: Charged Aerosol Detector

| Time (min) | Flow rate (mL/min) | %A | %B | %C |
|---|---|---|---|---|
| -4.0 | 0.8 | 1 | 64 | 35 |
| -0.5 | 0.8 | 1 | 64 | 35 |
| -0.2 | 0.2 | 1 | 64 | 35 |
| 0.0 | 0.2 | 1 | 64 | 35 |
| 0.1 | 0.8 | 1 | 64 | 35 |
| 2.0 | 0.8 | 4 | 61 | 35 |
| 7.0 | 0.8 | 10 | 60 | 30 |
| 13 | 0.8 | 10 | 60 | 30 |
| 14 | 0.8 | 1 | 64 | 35 |

| Phospholipid (PL) | Literature (PL-%) | Found (PL-%) |
|---|---|---|
| Phosphatidyl-Ethanolamine (PE) | 19.86 | 18.9 |
| Phosphatidyl-Inositol (PI) | 1.73 | 2.22 |
| Phosphatidyl-Serine (PS) | 0.0 | 0.0 |
| Phosphatidyl-Choline (PC) | 75.40 | 75.6 |
| LysoPhosphatidyl-Choline (LPC) | 0.04 | 0.92 |
| Total (mg/g) | 97.03 | 86.6 |




**Overlays of phospholipid standards (5000 ng o.c.) in triplicate**




**Analysis of egg yolk extract**

Download Poster Note: Analysis of Phospholipids in Natural Samples by Normal Phase HPLC and Corona Charged Aerosol Detection

thermoscientific

# Zwitterionic surfactants

Table of contents



## Lecithin (egg)

Methods for the measurement of triglycerides and phospholipids.

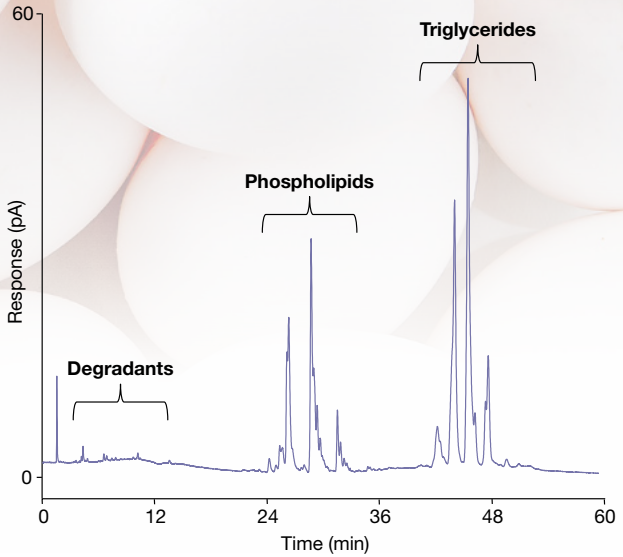
| | |
|---|---|
| HPLC column: | Acclaim C30, 5.0 µm, 4.6 × 150 mm |
| Column temperature: | 40 °C |
| Mobile phase A: | Acetonitrile |
| Mobile phase B: | Isopropanol |
| Mobile phase C: | 100 mM Ammonium Acetate, pH 5.0 |
| Flow rate: | 1.0 mL/min |
| Gradient: | See tables |
| Injection volume: | 2 µL |
| Detection: | Charged Aerosol Detector |

**Gradient for upper figure**

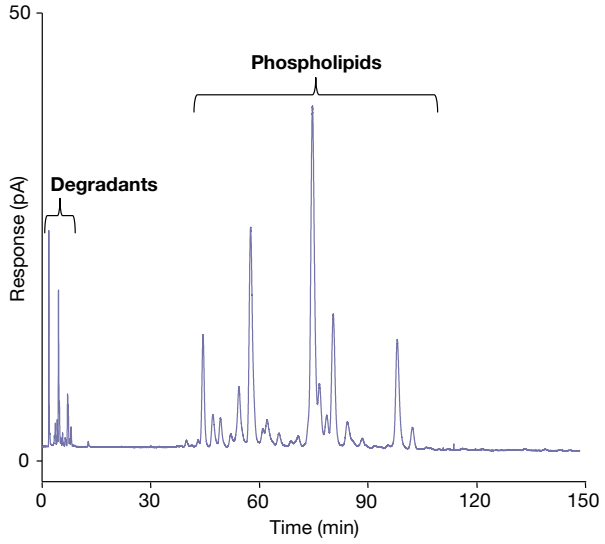
| Time (min) | %A | %B | %C |
|---|---|---|---|
| -15 | 70 | 0 | 30 |
| 0 | 70 | 0 | 30 |
| 0.1 | 70 | 0 | 30 |
| 10 | 90 | 0 | 10 |
| 35 | 10 | 80 | 10 |
| 50 | 0 | 95 | 5 |
| 60 | 0 | 95 | 5 |

**Gradient for lower figure**

| Time (min) | %A | %B | %C |
|---|---|---|---|
| -15 | 60 | 20 | 20 |
| 0 | 60 | 20 | 20 |
| 0.1 | 60 | 20 | 20 |
| 10 | 90 | 0 | 10 |
| 150 | 38 | 50 | 12 |
| 151 | 5 | 90 | 5 |
| 160 | 5 | 90 | 5 |



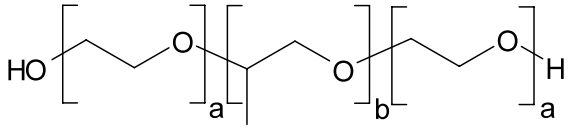

**Analysis of phospholipids and triglycerides in egg lecithin – commercial grade (PL30S)**




**Profiling Phospholipids in Egg lecithin – Commercial Grade (PL100M)**

Download Poster Note: Use of C30 as a general-purpose stationary phase for a broad range of applications

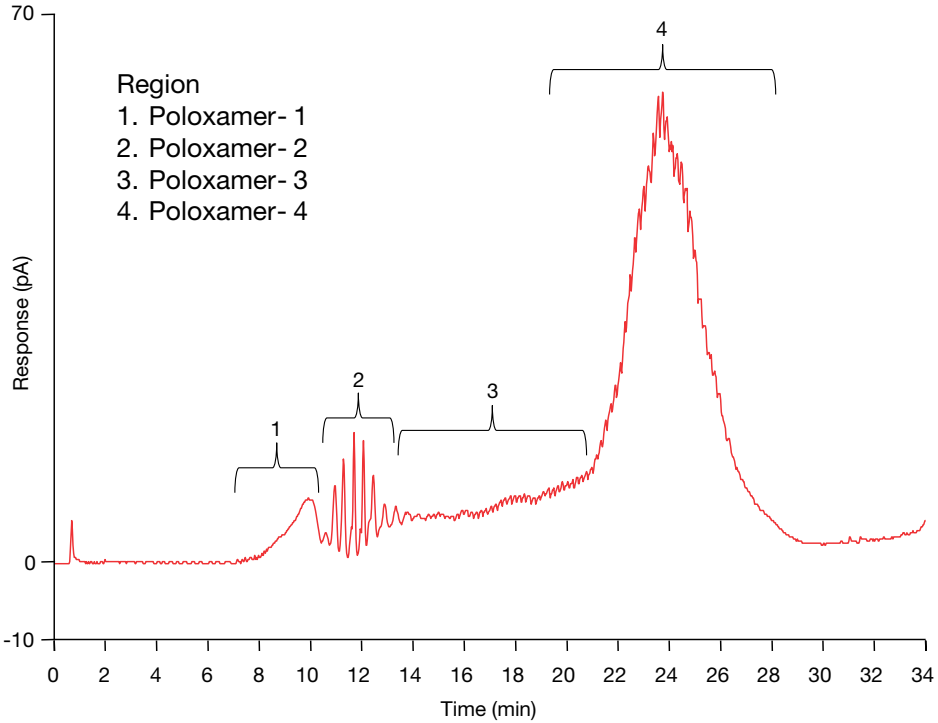
## Poloxamers




| | |
|---|---|
| HPLC column: | Accucore Vanquish C18, 1.5 µm, 2.1 × 150 mm |
| Column temperature: | 50 °C |
| Mobile phase A: | Aqueous Acetonitrile 50% (v/v) |
| Mobile phase B: | Tetrahydrofuran* |
| Flow rate: | 0.4 mL/min |
| Gradient: | See table |
| Injection volume: | 2 µL |
| Detection: | Charged Aerosol Detector |

| Time (min) | %A | %B |
|---|---|---|
| -6 | 100 | 0 |
| 0 | 100 | 0 |
| 3 | 100 | 0 |
| 25 | 50 | 50 |
| 33 | 10 | 90 |
| 34 | 100 | 0 |

*BHT free




**Characterization of poloxamer 407 (Pluronic F127)**

See Poster Note: Characterization and Lot-to-Lot Variability of Complex Surfactants by High Performance Liquid Chromatography and Charged Aerosol Detection

thermo scientific

# Nonionic surfactants

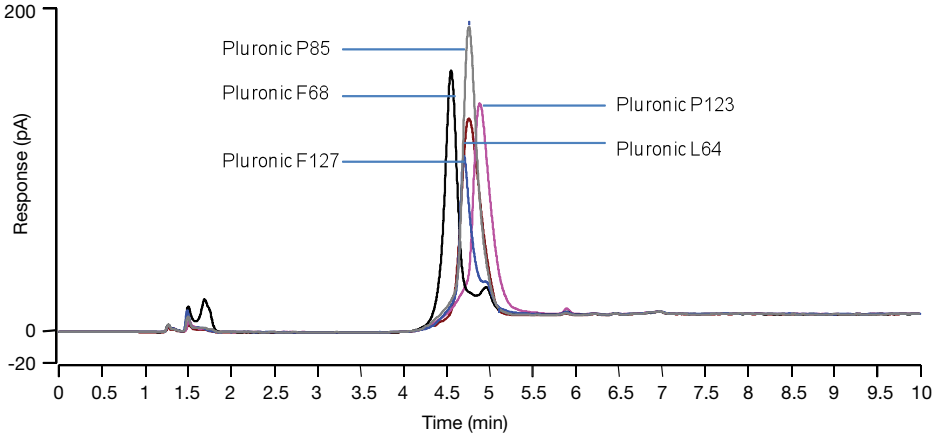
## Table of contents



## Poloxamers

HPLC column: Accucore Vanquish C18, 1.5 µm, 2.1 × 150 mm
Column temperature: 40 °C
Mobile phase A: 100 mM Ammonium Formate, pH 4.0
Mobile phase B: n-Propanol
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 40 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B | Curve |
|---|---|---|---|
| -5 | 95 | 5 | 5 |
| 0 | 95 | 5 | 5 |
| 1 | 95 | 5 | 5 |
| 7 | 50 | 50 | 2 |
| 10 | 10 | 90 | 5 |
| 10 | 10 | 90 | 5 |




**Analysis of different poloxamers. Each elutes as a single peak for more sensitive quantitation**

See Poster Note: Quantitation of Pluronics by High Performance Liquid Chromatography and Corona Charged Aerosol Detection

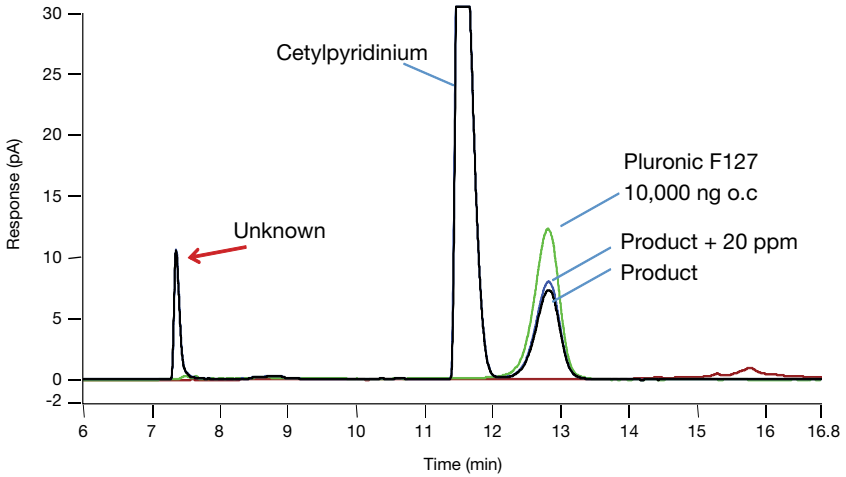
# Nonionic surfactants



## Poloxamer in mouthwash

HPLC column: Accucore Phenyl-1, 3 µm, 2.1 × 150 mm
Column temperature: 50 °C
Mobile phase A: 100 mM Ammonium Formate, pH 4.0
Mobile phase B: n-Propanol/Acetone 1:1 (v/v)
Flow rate: 0.4 mL/min
Gradient: See table
Injection volume: 20 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B | Curve |
|---|---|---|---|
| -5 | 95 | 5 | 5 |
| 0 | 95 | 5 | 5 |
| 1 | 95 | 5 | 5 |
| 7 | 50 | 50 | 2 |
| 10 | 10 | 90 | 5 |
| 10 | 10 | 90 | 5 |
| 19 | 10 | 90 | 5 |
| 19 | 95 | 5 | 5 |




HPLC chromatogram overlay of over-the-counter fluoride mouth rinse product, spiked and unspiked, and the poloxamer 407 standard at 10,000 ng o.c.

| Sample | Theoretical amount (ng o.c., ppm) | Experimental amount found (ng o.c., ppm) | Spike recovery (%) |
|---|---|---|---|
| Fluoride mouth rinse product | — | 5894, 294.7 | — |
| Spiked + 10 ppm | 6094, 304.7 | 6123, 306.2 | 114.5 |
| Spiked + 20 ppm | 6294, 314.7 | 6342, 317.1 | 112.0 |

Spike recovery results for poloxamer 407 in fluoride mouth rinse at two spike values.

See Poster Note: Quantitation of Pluronics by High Performance Liquid Chromatography and Corona Charged Aerosol Detection

thermoscientific

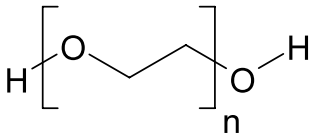
# Nonionic surfactants

## Table of contents

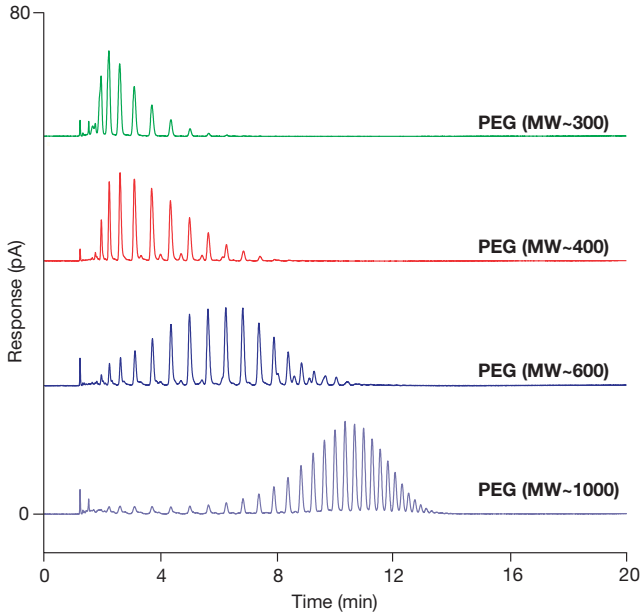


## Polyethylene glycol (PEG)

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm
Column temperature: 30 °C
Mobile phase A: 100 mM Ammonium Acetate, pH 5.0
Mobile phase B: Acetonitrile
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 5 µL
Detection: Charged Aerosol Detector




| Time (min) | %A | %B |
|---|---|---|
| -8 | 98 | 2 |
| 0 | 98 | 2 |
| 20 | 80 | 20 |




**Characterization of different PEGs**

Learn more: HPLC charged aersol detector analysis of polyethylene glycol (PEG)

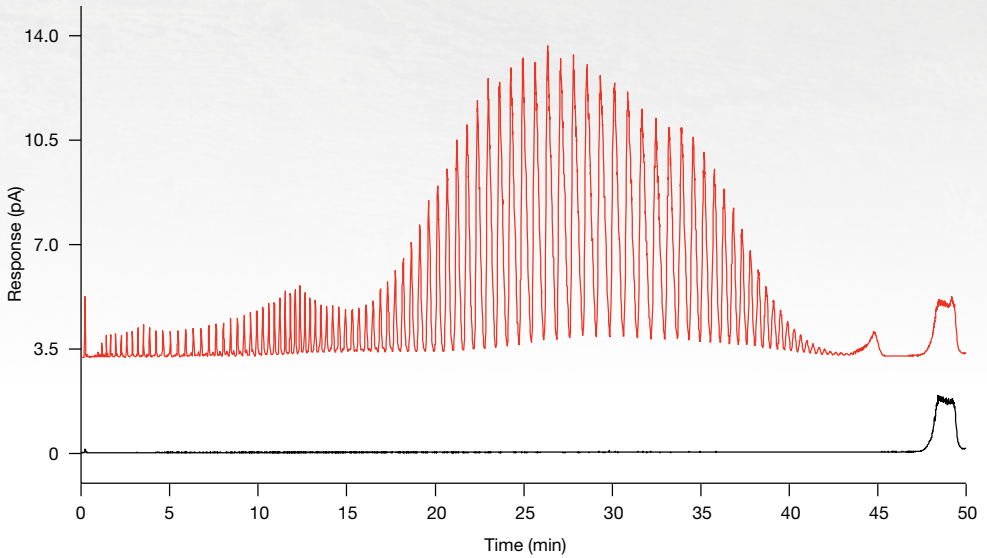
# Nonionic surfactants



## Polyethylene glycol – PEG 3000

HPLC column: UHPLC C18, 1.7 µm, 2.1 × 50 mm
Column temperature: 40 °C
Mobile phase A: DI Water
Mobile phase B: Acetonitrile
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 20 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| 0 | 94 | 6 |
| 1 | 88 | 12 |
| 3 | 83 | 17 |
| 12 | 74 | 26 |
| 17 | 72.5 | 27.5 |
| 25 | 71.5 | 28.5 |
| 32 | 70.5 | 29.5 |
| 43 | 67 | 33 |
| 47 | 50 | 50 |
| 48 | 20 | 80 |
| 49 | 20 | 80 |
| 50 | 94 | 6 |




**Characterization of PEG 3000. Red - standard (29 µg on column); black – distilled water blank**

Learn more: HPLC charged aersol detector analysis of polyethylene glycol (PEG-3000)

# Nonionic surfactants



## PEGylation

HPLC column: Thermo Scientific™ MAbPac™ RP, 4.0 µm, 3.0 × 100 mm

Column temperature: 60 °C, still air mode

Mobile phase A: 0.1% TFA in DI Water

Mobile phase B: 0.1% TFA in Acetonitrile

Flow rate: 0.6 mL/min

Gradient: See table

Injection volume: 1 µL

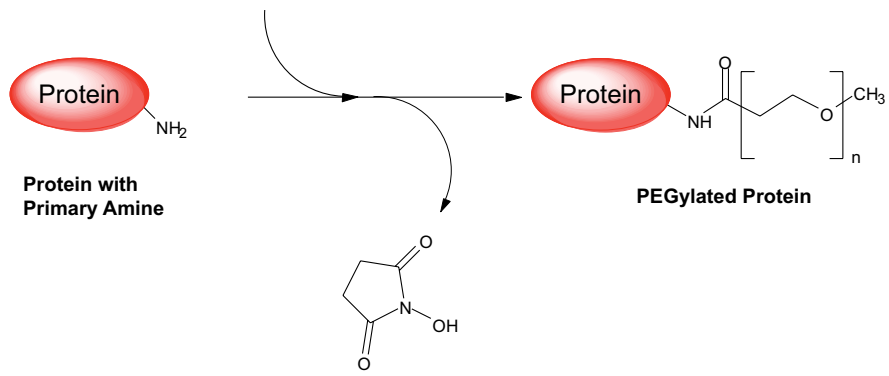
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| -5 | 90 | 10 |
| 0 | 90 | 10 |
| 10 | 60 | 40 |
| 12 | 10 | 90 |
| 15 | 10 | 90 |

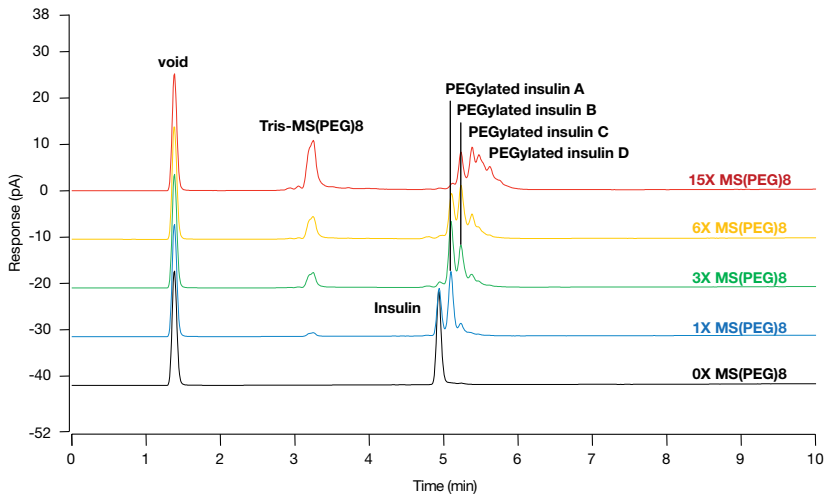
The HPLC-UV-CAD approach is able to quantify free PEG, partially PEGylated intermediates, and final PEGylated peptide/protein to optimize reaction conditions, assess the quality of final product, and study drug product stability.



**Methyl-PEG$_n$-NHS Ester**

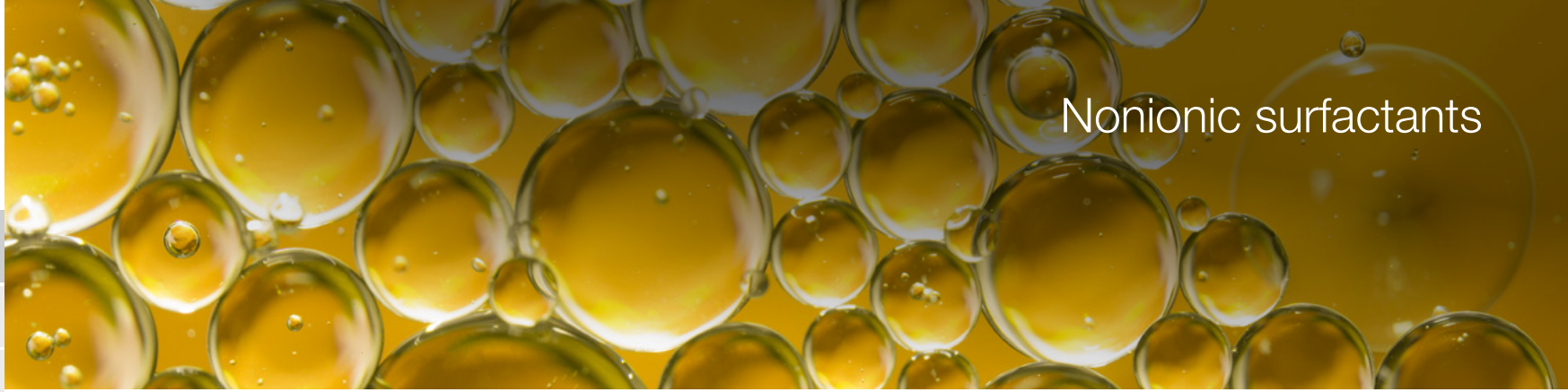



**Reaction of The PEGylation Agent MS(PEG)8 with the primary amine of a peptide or protein**




**Insulin PEGylation reactions monitored by UHPLC-UV-CAD**

See Poster Note: Monitoring Peptide PEGylation by HPLC with Charged Aerosol Detection



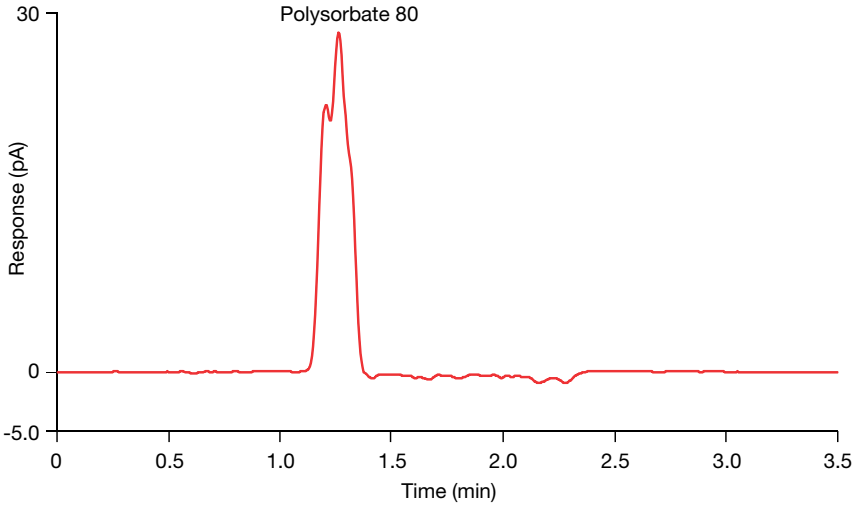
# Nonionic surfactants



## Polysorbates – quantification

| | |
|---|---|
| HPLC column: | Thermo Scientific™ Betasil™ C1, 5 µm, 3.0 × 100 mm |
| Column temperature: | 30 °C, still air mode |
| Mobile phase A: | Deionized water |
| Mobile phase B: | Acetonitrile/Methanol/Trifluoroacetic Acid (28/70/2) |
| Flow rate: | See table |
| Gradient: | See table |
| Injection volume: | 5 µL |
| Detection: | Charged Aerosol Detector |

| Time (min) | Flow Rate (mL/min) | %A | %B |
|---|---|---|---|
| 0 | 1.00 | 100 | 0 |
| 0.5 | 1.00 | 100 | 0 |
| 0.7 | 1.35 | 0 | 100 |
| 0.8 | 1.50 | 0 | 100 |
| 1.5 | 1.50 | 0 | 100 |
| 1.7 | 1.00 | 100 | 0 |
| 3.5 | 1.00 | 100 | 0 |



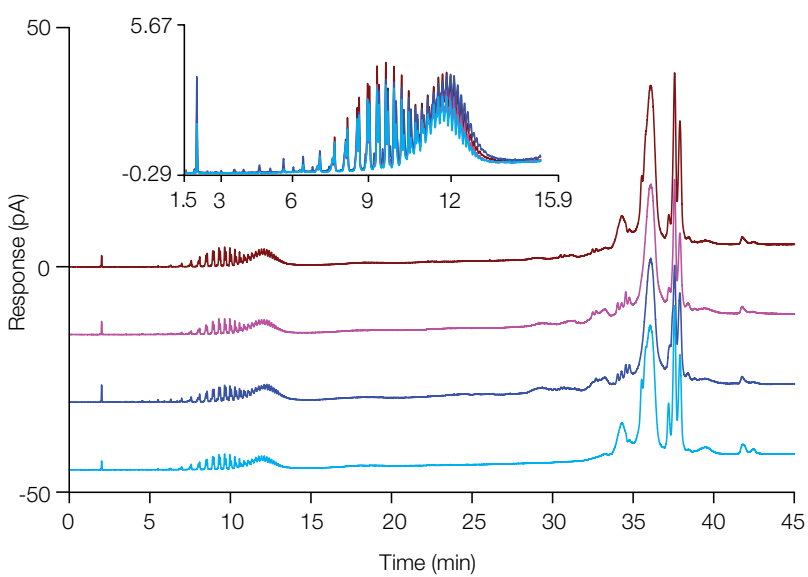

**Elution of polysorbate 80 standard as a single peak for more sensitive quantitation**

See Poster Note: Application of Charged Aerosol HPLC Detection in Biopharmaceutical Analysis

## Polysorbates – Material Characterization

| | |
|---|---|
| HPLC column: | Acclaim 300 C18, 3 µm, 4.6 × 150 mm |
| Column temperature: | 30 °C |
| Mobile phase A: | Acetonitrile/Methanol/DI Water/Trifluoroacetic Acid (8/2/90/0.1) |
| Mobile phase B: | Acetonitrile/Methanol/DI Water/Trifluoroacetic Acid (72/18/10/0.1) |
| Flow rate: | 0.4 mL/min |
| Gradient: | See table |
| Injection volume: | 2 µL |
| Detection: | Charged Aerosol Detector |

| Time (min) | %A | %B |
|---|---|---|
| 0 | 100 | 0 |
| 1 | 100 | 0 |
| 33 | 0 | 100 |
| 57 | 0 | 100 |
| 59 | 0 | 100 |
| 64 | 100 | 0 |



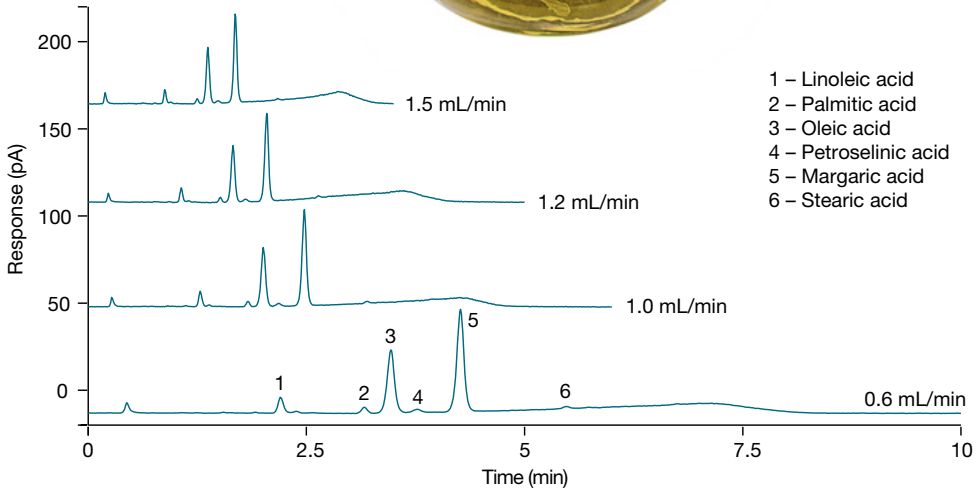

**Determination of lot-to-lot variability.** Stacked plot of four commercially available polysorbate 80 products. (Inset - low molecular weight components overlaid).

Download Poster Note: Evaluation of Methods for the Characterization and Quantification of Polysorbates and Impurities along with other Surfactants and Emulsifiers used in the Food and Pharmaceutical Industries

## Polysorbates – Polysorbate 80 fatty acid analysis

HPLC column: Core-shell C18, 2.6 µm, 2.1 × 100 mm
Column temperature: 25 °C, still air mode
Mobile phase A: 0.05% (v/v) Formic Acid in DI Water
Mobile phase B: 0.05% (v/v) Formic Acid in Acetonitrile
Flow rate: 0.6–1.5 mL/min
Gradient: See table
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| 0 | 25 | 75 |
| 0.8 | 25 | 75 |
| 2.5 | 15 | 85 |
| 3.0 | 15 | 85 |
| 3.5 | 25 | 75 |
| 4.5 | 25 | 75 |




*We are grateful to Professor Dr. U. Holzgrabe (University of Würzburg) for providing this figure.*

**Chromatographic separation of different fatty acids found in polysorbate 80 hydrolysates using different flow rates. 1) linoleic acid; 2) palmitic acid; 3) oleic acid; 4) petroselinic acid; 5) margaric acid; and 6) steric acid**

See Poster Note: Fatty Acid Analysis in Polysorbate 80 by UHPLC-CAD
Refer to article: Influence of charged aerosol detector instrument setting on the ultra-high-performance liquid chromatography analysis of fatty acids in polysorbate 80

# Nonionic surfactants

## Table of contents

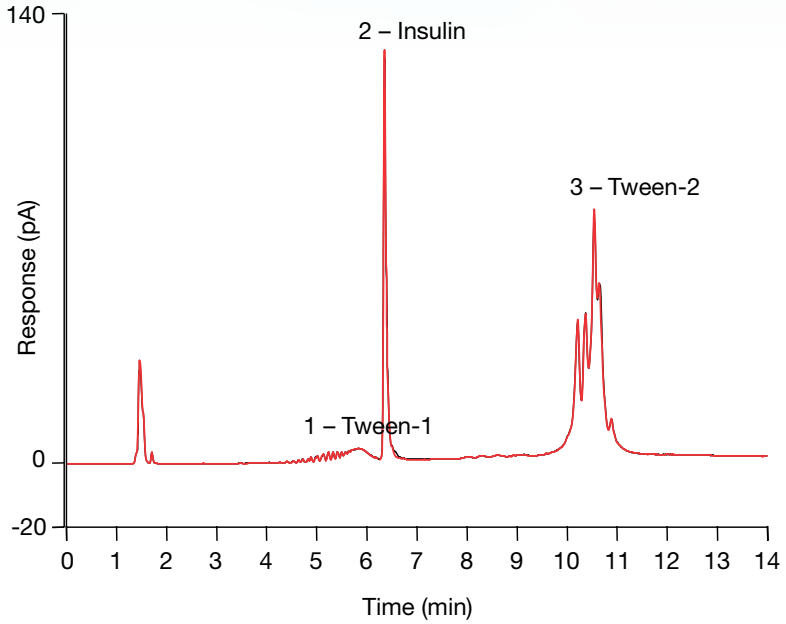


## Polysorbates – Polysorbate 80 and insulin in a biopharmaceutical formulation

| | |
|---|---|
| HPLC column: | Accucore 150-C4, 2.6 µm, 3 × 100 mm |
| Column temperature: | 50 °C |
| Mobile phase A: | 0.1% TFA |
| Mobile phase B: | Acetonitrile, 0.1 v/v-% Trifluoroacetic Acid |
| Mobile phase C: | n-Propanol/Tetrahydrofuran* (80:20) |
| Flow rate: | 0.5 mL/min |
| Gradient: | See table |
| Injection volume: | 20 µL |
| Detection: | Charged Aerosol Detector |

| Time (min) | %A | %B | %C |
|---|---|---|---|
| -5 | 90 | 10 | 0 |
| 0 | 90 | 10 | 0 |
| 7 | 25 | 75 | 0 |
| 7.1 | 0 | 0 | 100 |
| 12 | 0 | 0 | 100 |
| 13 | 0 | 100 | 0 |
| 14 | 90 | 10 | 0 |

*BHT free




**Simultaneous determination of peptide and surfactant in biopharmaceutical formulation**

For more information: HPLC-CAD analysis of polysorbate 80 and insulin in a biopharmaceutical formulation

# Nonionic surfactants

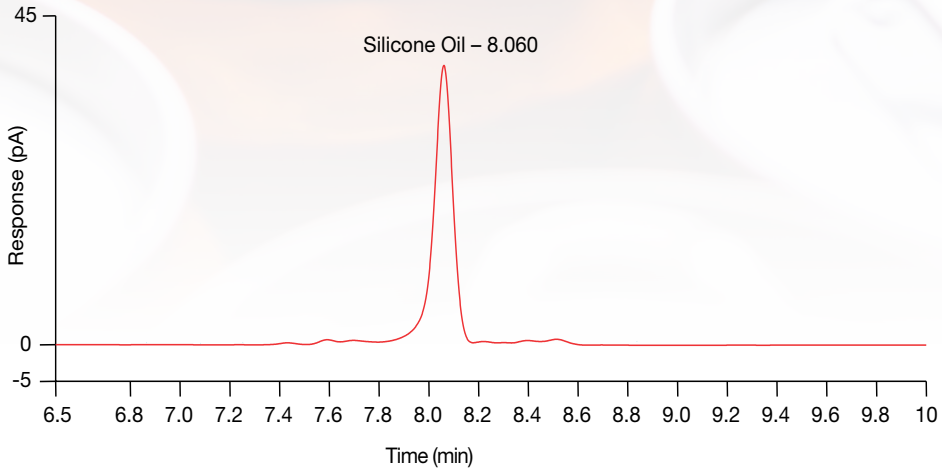


## Silicones – quantitation

HPLC column: Accucore C18, 2.6 µm, 3.0 × 150 mm
Column temperature: 40 °C
Mobile phase A: 0.5% Formic Acid/Acetonitrile/Tetrahydrofuran* (35:35:30)
Mobile phase B: Tetrahydrofuran*
Flow rate: See table
Gradient: See table
Injection volume: 2–10 µL
Detection: Charged Aerosol Detector

| Time (min) | Flow Rate (mL/min) | %A | %B |
|---|---|---|---|
| -5.0 | 0.6 | 100 | 0 |
| -0.2 | 0.6 | 100 | 0 |
| -0.1 | 0.3 | 100 | 0 |
| 0.0 | 0.3 | 100 | 0 |
| 0.5 | 0.6 | 100 | 0 |
| 4.0 | 0.6 | 100 | 0 |
| 6.0 | 0.6 | 0 | 100 |
| 8.0 | 0.6 | 0 | 100 |
| 10.0 | 1.0 | 0 | 100 |
| 12.0 | 1.0 | 50 | 50 |
| 14.0 | 0.6 | 100 | 0 |

*BHT free




**Elution of silicone standard as a single peak for more sensitive quantitation**

See Poster Note: Analysis of Silicone Oils by High Performance Liquid Chromatography and Corona Charged Aerosol Detection

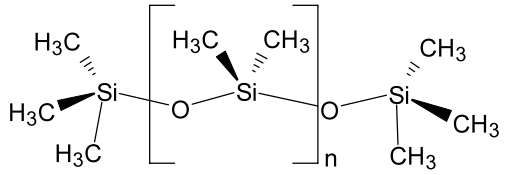
# Nonionic surfactants

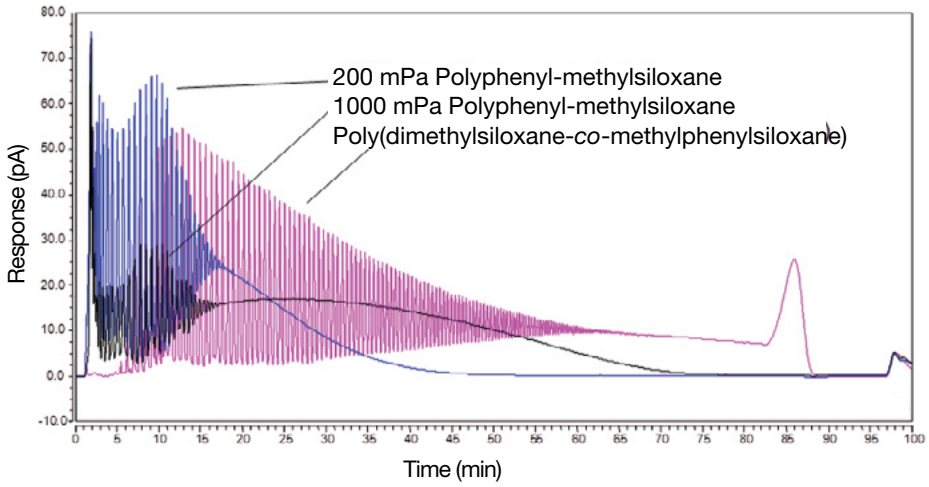


## Silicones – profiling

HPLC column: Accucore C18, 2.6 µm, 3.0 × 150 mm
Column temperature: 40 °C
Mobile phase A: Methanol
Mobile phase B: n-Propanol
Flow rate: 0.5 mL/min
Gradient: See table
Injection volume: 2–10 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B | Curve |
|---|---|---|---|
| -10 | 100 | 10 | 5 |
| 0 | 100 | 10 | 5 |
| 80 | 15 | 85 | 3 |
| 85 | 0 | 100 | 5 |
| 95 | 0 | 100 | 5 |
| 95 | 100 | 0 | 5 |
| 100 | 100 | 0 | 5 |




**Characterization of different silicones**

See Poster Note: Analysis of Silicone Oils by High Performance Liquid Chromatography and Corona Charged Aerosol Detection

# Nonionic surfactants



## SPAN – profiling

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm

Column temperature: 30 °C

Mobile phase A: 100 mM Ammonium Acetate, pH 5.4

Mobile phase B: Acetonitrile/Methanol/Tetrahydrofuran/Acetic Acid (500:375:125:4)

Flow rate: 0.8 mL/min

Gradient: See table

Injection volume: 10 µL

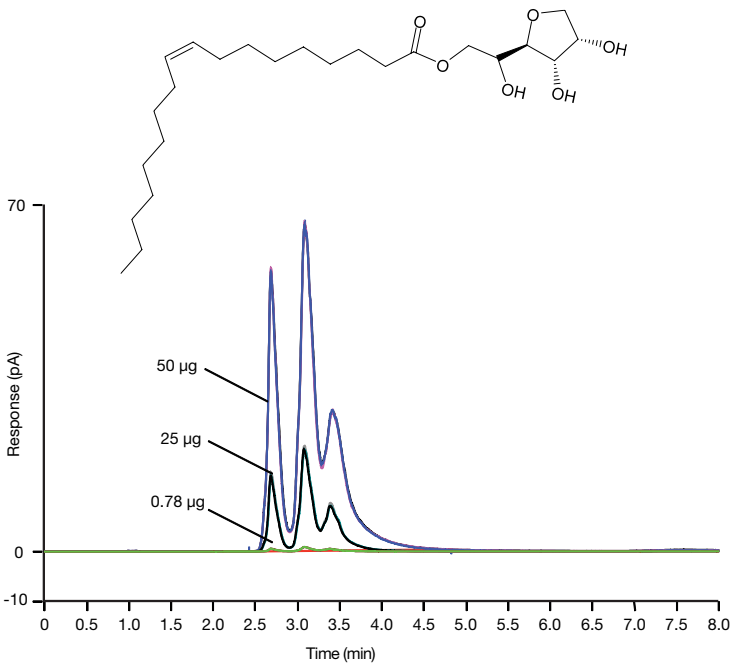
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| -5 | 90 | 10 |
| 0 | 90 | 10 |
| 5 | 10 | 90 |
| 8 | 5 | 95 |
| 8 | 90 | 10 |

| Compound | Span # | E# | FDA-listed (USA)* |
|---|---|---|---|
| Sorbitan monolaurate | 20 | 493 | |
| Sorbitan monopalmitate | 40 | 495 | |
| Sorbitan monostearate | 60 | 491 | ✓ |
| Sorbitan tristearate | 65 | 492 | |
| Sorbitan monooleate | 80 | 494 | ✓ |
| Sorbitan sesquioleate | 83 | – | |
| Sorbitan trioleate | 85 | – | |

Spans approved for use in foods and beverages

*Substances Added to Food (FDA)




**Analysis of Span 80 standard (each level in triplicate)**

For more information: HPLC charged aerosol detector analysis of sorbitan monooleate (Span 80)

## Triton X-100: quantitation

HPLC column: Betasil C1, 5 µm, 3.0 × 100 mm

Column temperature: 30 °C, still air mode

Mobile phase A: Deionized water

Mobile phase B: Acetonitrile/Methanol/Trifluoroacetic Acid (28/70/2)

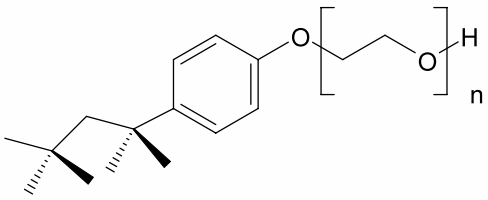
Flow rate: See table

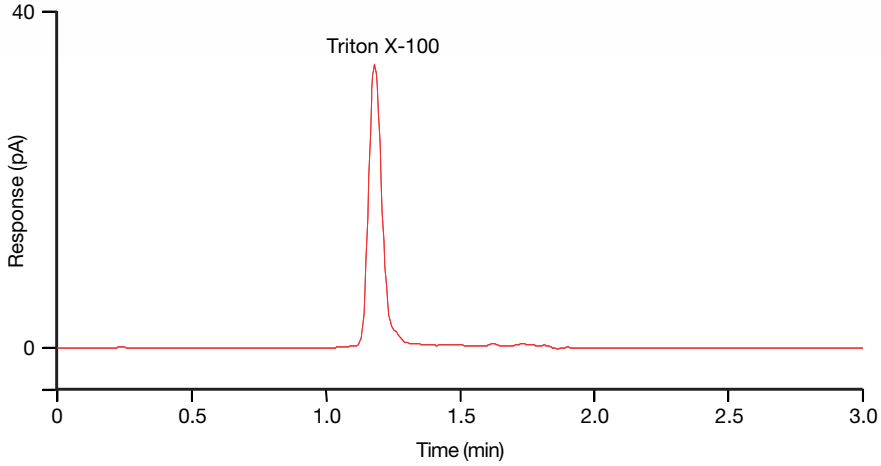
Gradient: See table

Injection volume: 5 µL

Detection: Charged Aerosol Detector

| Time (min) | Flow Rate (mL/min) | %A | %B |
|---|---|---|---|
| 0 | 1.00 | 100 | 0 |
| 0.5 | 1.00 | 100 | 0 |
| 0.7 | 1.35 | 0 | 100 |
| 0.8 | 1.50 | 0 | 100 |
| 1.5 | 1.50 | 0 | 100 |
| 1.7 | 1.00 | 100 | 0 |
| 3.5 | 1.00 | 100 | 0 |




**Elution of Triton X-100 standard as a single peak for more sensitive quantitation**

See Poster Note: Application of Charged Aerosol HPLC Detection in Biopharmaceutical Analysis

thermo scientific

# Nonionic surfactants

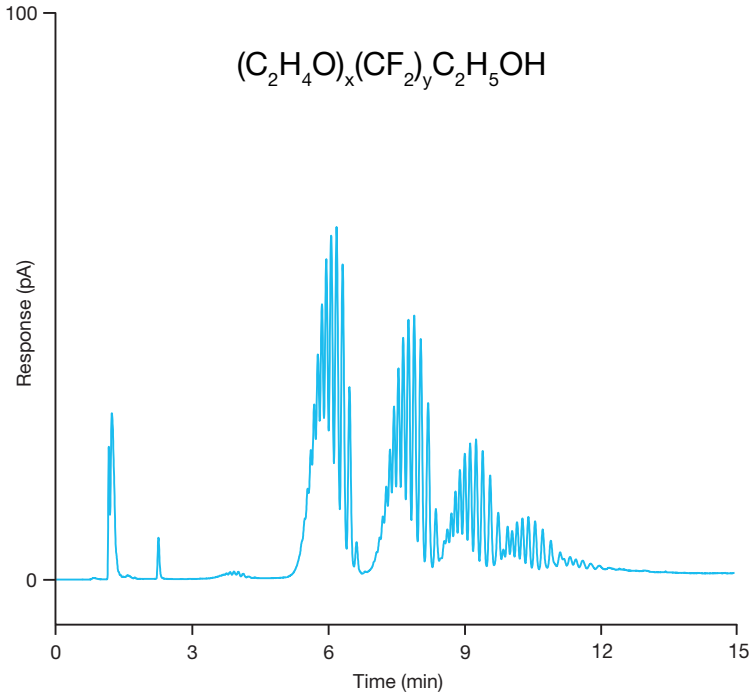
## Table of contents



## Zonyl® FSO Fluorosurfactant

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm
Column temperature: 30 °C
Mobile phase A: 100 mM Ammonium Acetate, pH 5.2
Mobile phase B: Acetonitrile
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 5 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| -8 | 70 | 30 |
| 0 | 70 | 30 |
| 10 | 40 | 60 |
| 15 | 40 | 60 |




**Characterization of Zonyl FSO Fluorosurfactant**

For more information: Analysis of Zonyl FSO fluorosurfactant by HPLC-CAD

thermoscientific

# Multiple surfactants

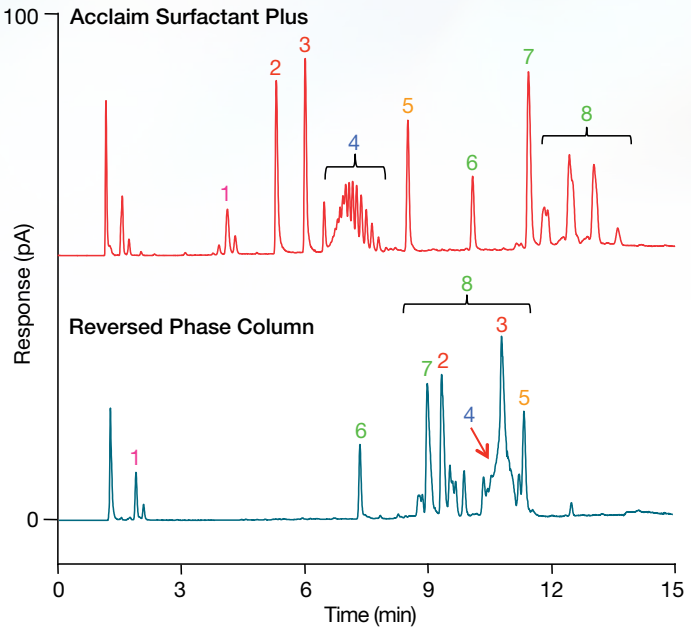
## Table of contents



The Acclaim Surfactant Plus column provides ideal selectivity for the simultaneous separation of anionic, nonionic, cationic, and amphoteric surfactants, whereas the C18 column fails to resolve them under the same or other conditions.

| | |
|---|---|
| HPLC column: | Acclaim Surfactant Plus, 3 µm, 3.0 × 150 mm |
| Column temperature: | 30 °C |
| Mobile phase A: | Acetonitrile |
| Mobile phase B: | 0.1 M Ammonium Acetate, pH 5 |
| Flow rate: | 0.60 mL/min |
| Gradient: | See table |
| Injection volume: | 5 µL |
| Detection: | Charged Aerosol Detector |

| Time (min) | %A | %B |
|---|---|---|
| -8 | 25 | 75 |
| 0 | 25 | 75 |
| 10 | 80 | 20 |
| 15 | 80 | 20 |




Peaks: (100–400 µg/mL each)

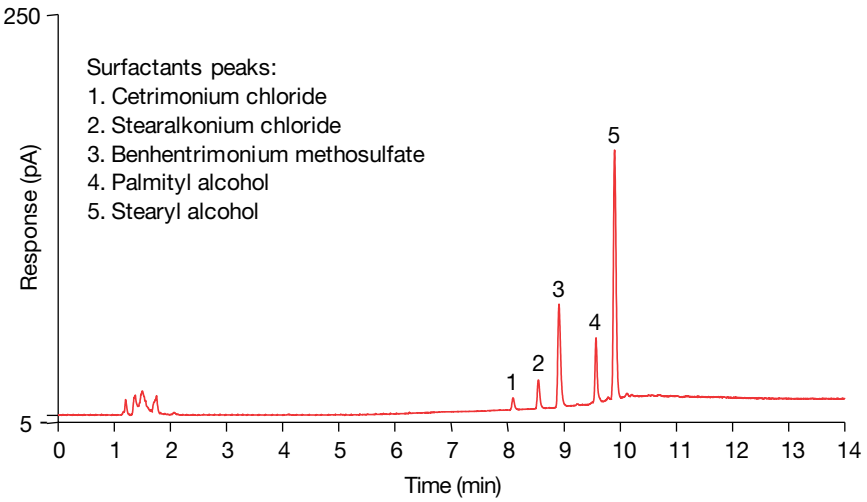
1. Xylene sulfonate
2. Laurylpyridinium
3. Lauryldimethylbenzyl ammonium
4. Triton X-100
5. Cetyl betaine
6. Decyl sulfate
7. Dodecyl sulfate
8. Linear alkylbenzene sulfonate (LAS)

Download the Poster Note: Exploring Mixed-Mode Chromatorgraphy: Column Chemistry, Properties, and Applications

# Multiple surfactants

HPLC column: Acclaim Surfactant Plus, 3 µm, 3.0 × 150 mm
Column temperature: 50 °C
Mobile phase A: 50 mM Ammonium Acetate, pH 5 in Acetonitrile/DI Water (1:1)
Mobile phase B: 50 mM Ammonium Acetate, pH 5 in Acetonitrile/Methanol/DI Water (5:4:1)
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 5 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| -5 | 95 | 5 |
| 0 | 95 | 5 |
| 1 | 95 | 5 |
| 7 | 50 | 50 |
| 10 | 10 | 90 |
| 10 | 10 | 90 |
| 14 | 10 | 90 |




Download the Poster Note: Quantitation of Surfactants by High Performance Liquid Chromatography and Corona Charged Aerosol Detection

thermoscientific

# Multiple surfactants

## Table of contents



## Laundry detergent

HPLC column: Acclaim Surfactant Plus, 3 µm, 4.6 × 250 mm

Column temperature: 30 °C

Mobile phase A: 50 mM Ammonium Acetate, pH 5 in DI Water/Acetonitrile (9:1)

Mobile phase B: 50 mM Ammonium Acetate, pH 5 in Acetonitrile/Methanol/DI Water (4:5:1)

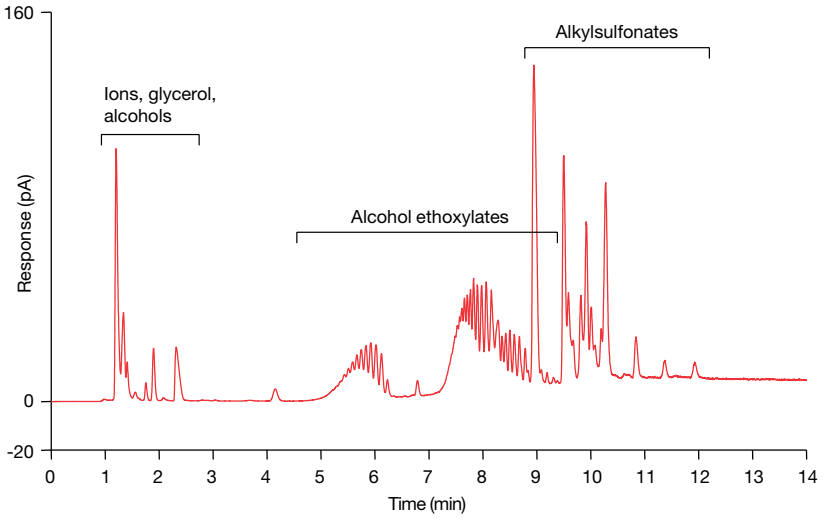
Flow Rate: 0.6 mL/min

Gradient: See table

Injection Volume: 5.0 µL

Detection: Charged Aerosol Detector

| Time (min) | %A | %B |
|---|---|---|
| -5 | 80 | 20 |
| 0 | 80 | 20 |
| 2 | 80 | 20 |
| 12 | 10 | 90 |
| 17 | 10 | 90 |




See Poster Note: Direct Analysis of Surfactants using HPLC with Charged Aerosol Detection

thermoscientific

# Multiple surfactants

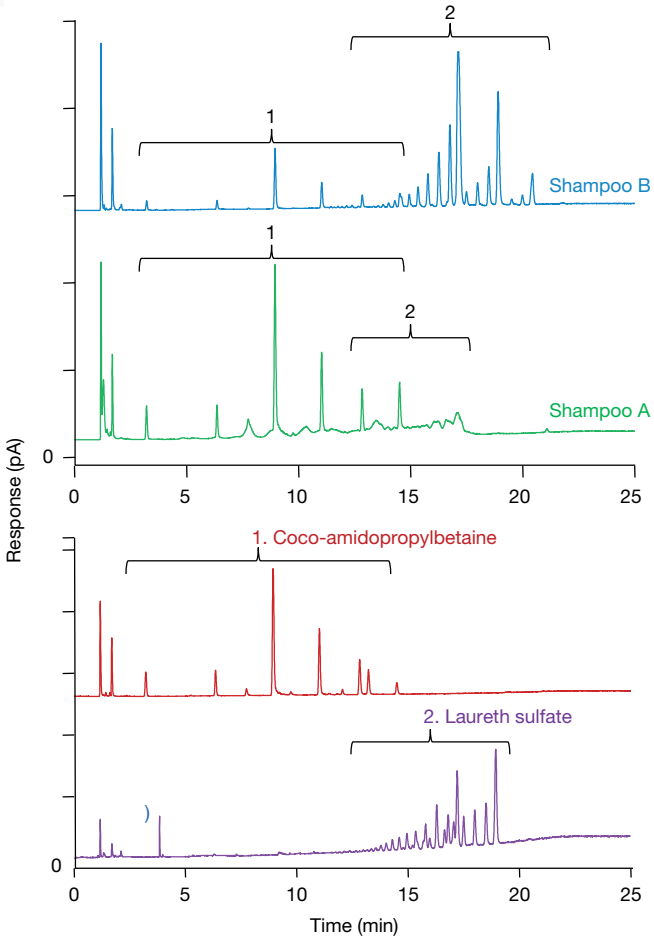
## Table of contents



## Shampoo

HPLC column: Acclaim Surfactant Plus, 3.0 µm, 3.0 × 150 mm
Column temperature: 25 °C
Mobile phase A: Acetonitrile
Mobile phase B: 100 mM Ammonium Acetate, pH 5
Mobile phase C DI Water
Flow rate: 0.6 mL/min
Gradient: See table
Injection volume: 1 µL
Detection: Charged Aerosol Detector

| Time (min) | %A | %B | %C |
|---|---|---|---|
| -7 | 20 | 50 | 30 |
| 0 | 20 | 50 | 30 |
| 18 | 80 | 20 | 0 |
| 25 | 80 | 20 | 0 |




For more information: Improved analysis of laureth sulfate and coco-amidopropyl betaine in shampoos by HPLC-CAD

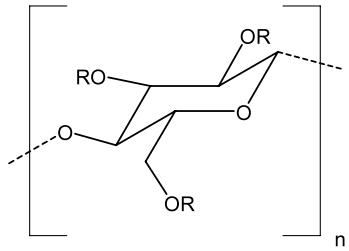
# Emulsifier



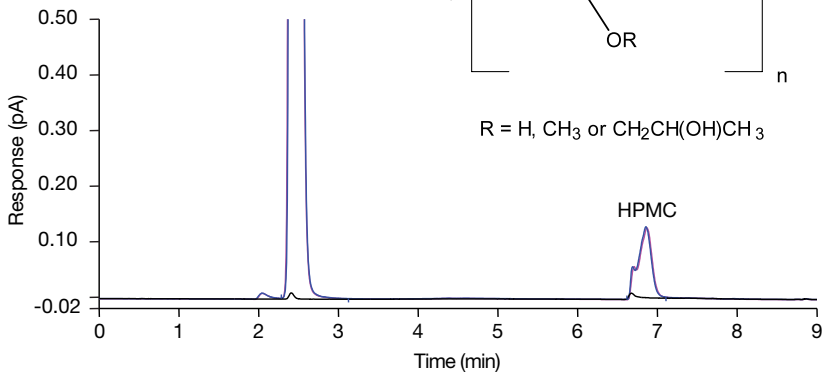
## Hypromellose (HPMC)

| | |
|---|---|
| HPLC column: | Non-porous C18, 2.0 µm, 4.6 × 150 mm |
| Column temperature: | 40 °C |
| Mobile phase A: | Deionized Water |
| Mobile phase B: | Acetonitrile |
| Mobile phase C: | Isopropanol |
| Gradient: | See poster note |
| Flow rate: | 0.4 mL/min |
| Injection volume: | 2–10 µL |
| Detection: | Charged Aerosol Detector |

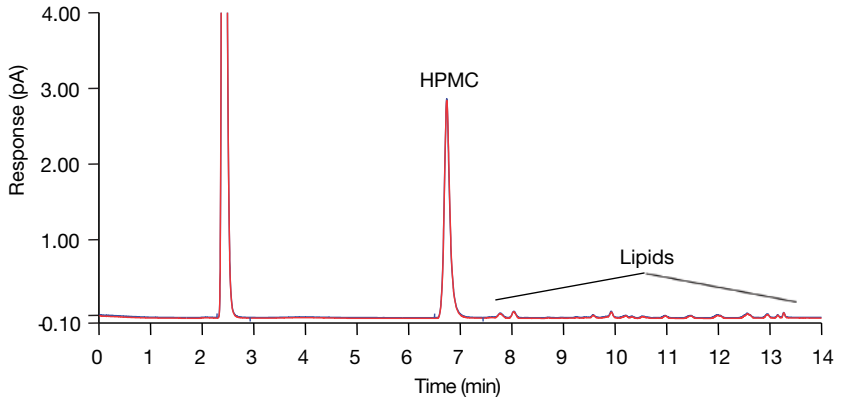
| Sample | HPMC found (mass-%) | Claimed Amount |
|---|---|---|
| Popsicle | 0.05 | <1% |
| Dairy Product | 0.21 | <1% |




R = H, $CH_3$ or $CH_2CH(OH)CH_3$




**Measurement of HPMC in a popsicle**




**Measurement of HPMC in a dairy product using extended gradient**

See Poster Note: Analysis of Emulsifiers in Foods by HPLC and Corona Charged Aerosol Detection



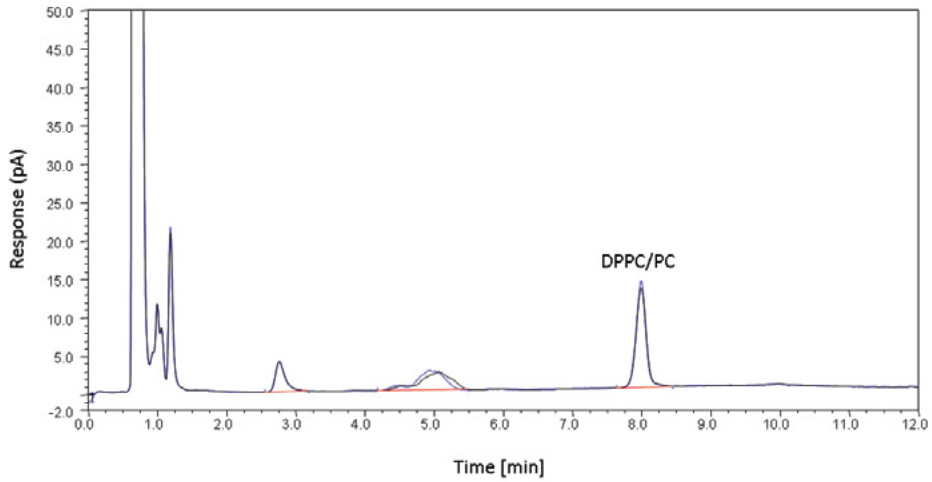
## Lecithin

HPLC column: Hypersil silica, 5 µm, 3.0 × 150 mm
Column temperature: 50 °C
Mobile phase A: DI Water
Mobile phase B: Isopropanol
Mobile phase C: Iso-octane
Flow rate: See table
Gradient: See table
Injection Volume: 2–10 µL
Detection: Charged Aerosol Detector

| Time (min) | Flow rate (mL/min) | %A | %B | %C |
|---|---|---|---|---|
| -2.0 | 1.5 | 2 | 63 | 35 |
| -0.5 | 1.5 | 2 | 63 | 35 |
| -0.2 | 0.2 | 2 | 63 | 35 |
| 0.0 | 0.2 | 2 | 63 | 35 |
| 0.2 | 1.5 | 7 | 58 | 35 |
| 7.0 | 1.5 | 11 | 54 | 35 |
| 11 | 1.5 | 11 | 54 | 35 |
| 12 | 1.5 | 2 | 63 | 35 |

This note presents a general method for the measurement of phosphatidylcholine (PC) as dipalmitoylphosphatidylcholine (DPPC), in various matrices. Results match the American Oil Chemists' official method (Ja 7c-07) for phospholipids.

| Sample | Phosphotidyl-choline (mass-%) | Claim amount | % of target |
|---|---|---|---|
| Lecithin, laboratory grade | 47.7 | N/A | N/A |
| Granola bar | 0.05 | <2% | N/A |
| Krill oil | 34.1 | 34.9 | 97.7 |




**Analysis of lecithin in a granola bar extract**

See Poster Note: Analysis of Emulsifiers in Foods by HPLC and Corona Charged Aerosol Detection

## Defoaming agent

A defoamer or an anti-foaming agent is a chemical additive that reduces and hinders the formation of foam. Commonly used agents are insoluble oils, polydimethylsiloxanes and other silicones, certain alcohols, stearates, and glycols. Anti-foaming agents have many practical uses: they are added to detergents to reduce foaming that might decrease the action of the detergent (e.g., dishwasher detergents have to be low foaming for the dishwasher to work properly); they are an ingredient in food (to curb effusion or effervescence in preparation or serving); or they are used in the cooking process (e.g., silicone oil is also added to cooking oil to prevent foaming in deep-frying). Defoamers are used in many industrial processes and products: paper, paint, industrial wastewater treatment, oil drilling, machine tool industry, oils, cutting tools, hydraulics, wood pulp, etc. Anti-foaming agents are also sold as over-the-counter medications (containing the drug simethicone) to relieve bloating.

## Detergent

A detergent contains a surfactant (or a mixture of surfactants) used for its cleaning properties. For aqueous-based systems, detergents emulsify oils, hold dirt in suspension, and act as wetting agents. Unlike soap, they do not form a scum with salts from hard water. Most commonly, the term “detergent” refers specifically to laundry detergent or dish detergent, as opposed to hand soap or other types of cleaning agents. The formulations can be complex, reflecting the diverse demands of the application and the highly competitive consumer market.

Some detergents are designed to be used in oil-based systems where they hold insoluble foreign matter in suspension, e.g., in lubricating oils and dry-cleaning solvents. Detergents are used to prevent carburetor and fuel injector fouling.

## Dispersant

A dispersant or a dispersing agent is either a non-surface active polymer or surfactant(s) added to a suspension to improve the separation of particles and to prevent settling or clumping. Dispersants maintain contaminants in suspension. Dispersants are used in many industries:

- They are added to automotive engine oils to prevent the accumulation of varnish like deposits on the cylinder walls, and to gasoline to prevent the buildup of gummy residues.
- They are used as an oil drilling aid to break up solids into fine particles, or liquids into droplets. As foam drilling fluids they are used to disperse foam bubbles into the air or gas.
- They can be used to dissipate oil slicks.
- They are used to prevent formation of biofouling or biofilms in industrial processes. It is also possible to disperse bacterial slime and increase the efficiency of biocides.

## Emulsifier

An emulsifier (also known as an "emulgent") is a substance that stabilizes an emulsion. One class of emulsifier is the surfactants. The terms surfactant and emulsifier are often used interchangeably. However, this tends to be industry specific with food chemists preferring to use emulsifier while industrial chemists favor the use of the term surfactant. Depending upon their structure, emulsifiers can show differences in their solubility in water and oil. Emulsifiers that are more soluble in water (and conversely, less soluble in oil) will generally form oil-in-water emulsions (e.g., crema [foam] in espresso, mayonnaise and hollandaise sauces, homogenized milk, vinaigrette) while emulsifiers that are more soluble in oil will form water-in-oil emulsions (e.g., butter and margarine). Examples of food emulsifiers include egg yolk (in which the main emulsifying agent is lecithin) and soy lecithin.

## Foaming agents

Some surfactants are foaming agents. When present in small amounts, they reduce the surface tension of a liquid thereby facilitating formation of a foam. These agents include sodium laureth sulfate (or sodium lauryl ether sulfate), sodium lauryl sulfate (or sodium dodecyl sulfate) and ammonium lauryl sulfate (ALS).

## Highly hydrophilic hydrotropes

A hydrotrope is a compound that solubilizes hydrophobic compounds in aqueous solutions. Examples include: sodium naphthalene sulfonate, xylene sulfonate, and sodium lauryl ether sulfate. Like surfactants, their structure consists of a hydrophilic part and a hydrophobic part, but the latter is generally too small to cause spontaneous self-aggregation. Highly hydrophilic hydrotropes enhance the solubility of other organics in water. Without them it would be impossible to incorporate sufficient quantities of surfactants, phosphates and solvents into detergent formulations.

## Hypromellose

Hypromellose (E464) (hydroxypropyl methylcellulose [HPMC]), is a semisynthetic, inert polymer used as an emulsifier, thickening, and suspending agent. It has a wide variety of commercial uses. In foods, it is often used to thicken dairy products and help improve flavor characteristics. It is used as an alternative to animal gelatin. In the pharmaceutical industry it is used as an excipient, control release agent, binder, and in coatings. Other uses include in construction materials (tile adhesives, cement renders, paints, and coatings), cosmetics, detergents and cleaners, and ophthalmic applications (eye drops, contact lenses).

## Laureth sulfate

Laureth sulfate (alkyl polyethoxy sulfate or lauryl ether sulfate), not to be confused with lauryl sulfate (sodium dodecyl sulfate), is a popular class of surfactants featuring good water solubility, high foam, and good cleaning properties. Sodium laureth sulfates (SLES) are used in many personal care products such as shampoo and hand soap. Different grades feature various mixtures of C12, C14, and C16 alkyl chains with varying degrees of ethoxylation.

## Lecithin

Lecithin (E322) is a general term used to describe any group of yellow brownish fatty substances occurring in animal and plant tissues and composed of a number of lipids (primarily phospholipids, e.g., phosphatidylcholine [PC], phosphatidylethanolamine [PE], and phosphatidylinositol [PI]). The composition of lecithin depends on the source, e.g., soybean lecithin contains 33–35% soybean oil, 19–21% PC, 8-20% PE and 20–21% PI. Lecithin is sometimes used to describe extracts high in PC.

Lecithin is used commercially in foods requiring a natural emulsifier or lubricant. It is commonly used as an emulsifier in chocolate and spray oils to prevent sticking. It promotes the homogeneous mixing of ingredients and improves shelf life for some products (e.g., it keeps cocoa and cocoa butter in candy from separating). In emulsions and fat spreads, it stabilizes the product and improves the texture of spreads.

## PEGylation

It is an increasingly common practice to modify biotherapeutic peptides, proteins, and small molecules by covalently attaching chemical groups designed to enhance properties such as bioavailability, potency, or stability. One example of this is the PEGylation reaction that attaches hydrophilic polyethylene glycol moieties tailored to increase the drug's aqueous solubility and circulating half life. Chemists developing extended-life biotherapeutics need to measure free PEG, partially PEGylated intermediates, and final PEGylated peptide/protein to optimize reaction conditions, assess the quality of final product, and study drug product stability.

## Polyethylene glycol

Polyethylene glycol (INS1521) is a polyether compound – a polymer of ethylene glycol. A number of PEGs exist based on molecular weight and geometry.

PEG has many uses. In the food industry PEG is used as an anti-foaming agent, carrier, emulsifier, glazing agent, and thickener. PEG is used in cosmetics (e.g., skin creams), personal lubricants, and toothpastes. Macrogol is the international nonproprietary name for PEG when it is used in medicine, e.g., as laxatives. Macrogols are also attached to biopharmaceutical drugs (PEGylation) to slow down their degradation, increase their duration of action, and reduce immunogenicity.

## Poloxamers

These are nonionic surfactants. They are nonionic triblock copolymers composed of a central hydrophobic chain of polyoxypropylene flanked by two hydrophilic chains of polyoxyethylene. Poloxamers are known by the trade names Kolliphor, Pluronics, and Synperonics. Poloxamers are commonly used in industrial applications, cosmetics, and pharmaceuticals that have been evaluated for drug delivery applications.

## Polysorbates

Polysorbates are oily liquids derived from ethoxylated sorbitan (formed from sorbitol) esterified with fatty acids. The number following polysorbate refers to the type of fatty acid associated with the polyoxyethylene sorbitan part of the molecule. Monolaurate is indicated by 20, monopalmitate is indicated by 40, monostearate by 60 and monooleate by 80. Common brand names include Alkest®, Canarcel®, Scattics, and Tween®.

Polysorbates are nonionic surfactants and emulsifiers and are used in large quantities throughout the food, cosmetic, and pharmaceutical industries. For example, polysorbate 80 is used as an emulsifier in foods, particularly in ice cream making the product smoother and easier to handle, as well as increasing its resistance to melting. In biopharmaceuticals, it prevents surface adsorption and stabilizes proteins against stress-induced aggregation, such as agitation and shear.

Because polysorbates have a significant demand across industries they are often produced in large lots with varying limits of impurities (peroxides, carbonyls, and metals) reported. The characterization and quantification of polysorbates is difficult because these compounds are heterogeneous mixtures with no chromophore. As a result, physical tests and testing for impurities are typically used for release criteria. While this testing is sufficient for the manufacturers to release quality material, it may not be sufficient for the end user. Because polysorbates are used in final formulations, in-depth characterization and determination of lot-to-lot variability is critical to both the pharmaceutical and biopharmaceutical industries.

## siRNA

Short segments of RNA, called small interfering RNA (siRNA), are being investigated as novel approaches to treat numerous conditions including cancer, AIDS, diabetes, age-related macular degeneration, and hepatitis. To effectively penetrate the cell membrane, the RNA complex can be encapsulated with transfection reagents such as cationic lipid liposomes. As clinical trials proceed to final stages, quality control measurements of the purity and quantity of these cationic lipid delivery agents will be required.

## Silicones

Silicone oils are polymeric organosilicon compounds with diverse chemical structures and many practical uses. One silicone - Polydimethylsiloxane (PDMS) is used as an anti-foaming agent in cooking oil and in a number of processed foods, fast food items, and sodas. Silicones are used industrially as lubricants and hydraulic fluids, in dashpots, wet type transformers, diffusion pumps, and in oil-filled heaters. In consumer products PDMS, also called dimethicone, is used as an antiflatulent.

## Shampoo, shower gels and hair conditioners

Shampoo formulations use blends of surfactants to produce the desired qualities. Many contain laureth sulfate and coco-amidopropylbetaine. Anionic laureth sulfate surfactants feature good water solubility, high foam, and good cleaning properties; typically they are mixtures of C12, C14, and C16 alkyl chains with varying degrees of ethoxylation. Zwitterionic coco-amidopropylbetaine has moderate cleaning properties and low irritation to mucous membranes.

Hair conditioners primarily use cationic surfactants sometimes in combination with nonionic surfactants. The cationic surfactant is deposited onto the slightly negatively charged hair and acts as a lubricant.

## Spans

Sorbitan esters (also known as Spans) are nonionic surfactants that are derived from sorbitan – the precursor also used to produce polysorbates. A wide variety of Spans are commercially available .The number following a Span refers to the type of fatty acid associated with the sorbitan part of the molecule, e.g., Span 80 (sorbitan monooleate), Span 83 (sorbitan sesquioleate), and Span 85 (sorbitan trioleate).

Spans are used as emulsifying agents in the preparation of emulsions, creams and ointments for pharmaceutical, and cosmetic use. Spans are used as emulsifiers and stabilizers in food products. In the US, Span 60 and 80 are FDA-listed as food additives while in the EU the range is increased to include Span 20, 40, 60, 65, and 80.

Span is also an oil dispersant and is one of the components in Corexit® 9500 used to treat the oil spill in the Gulf of Mexico in 2013.

## Wetting agents

Wetting agents are substances that reduce the surface tension of water to allow it to spread more easily. Many technological processes require control of liquid spreading over solid surfaces. By reducing the surface tension with surfactants, a non-wetting material can be made to become partially or completely wetting. Wetting is important in cleaning applications. For example, dish soap is a wetting agent.

## Zonyl FSO fluorosurfactant

An ethoxylated nonionic fluorosurfactant that is sparingly water-soluble and gives extremely low aqueous surface tensions. As an additive to adhesives, paints, and coatings it provides superior weatherability, UV stability and resistance to soiling.

# Thermo Scientific references




| Title | Authors | Publication |
|---|---|---|
| Fatty Acid Analysis in Polysorbate 80 by UHPLC-CAD | Schilling, K; Pawellek, R; Lovejoy, K; Muellner, T; Holzgrabe, U. | n/a, 2018 |
| A Highly Sensitive High-Performance Liquid Chromatography-Charged Aerosol Detection Method for the Quantitative Analysis of Polysorbate 80 in Protein Solution | Long, Z; Shen, G; Neubauer, M; Lovejoy, K; Liu, L; Liu, X; Jin, Y; Liu, X | AN72398, 2017 |
| Characterization of a Biologic Therapeutic: Reversed Phase Analysis of Protein and Excipients | Bailey, B; Acworth. I; Pippitt, J; Steiner, F; Sneekes, E-J | PN72089, 2016 |
| Analytical Methods to Qualify and Quantify PEG and PEGylated Biopharmaceuticals | Crafts, C; Bailey, B; Plante, M; Acworth, I | AN1015, 2016 |
| Direct Analysis of Surfactants using HPLC with Charged Aerosol Detection | Plante, M; Bailey, B; Acworth, I; Crafts, C | PN70055, 2016 |
| Characterization and Lot-to-Lot Variability of Complex Surfactants by High Performance Liquid Chromatography and Charged Aerosol Detection | Plante, M; Acworth, I; Bailey, B; Sneekes, E-J; Steiner, F | PN64687, 2016 |
| Fast and Sensitive Determination of Quaternary Amines by UHPLC | Plante, M; Acworth, I; Bailey, B; Sneekes, E-J; Steiner, F | PN71688, 2016 |
| Quantitation of Pluronics by High Performance Liquid Chromatography and Corona Charged Aerosol Detection | Plante, M; Bailey, B; Acworth, I | PN70535, 2016 |
| Quantitation of Surfactants in Samples by high Performance Liquid Chromatography and Corona Charged Aerosol Detection | Plante, M; Bailey, B; Acworth, I | PN70539, 2016 |
| Analysis of Phospholipids in Natural Samples by Normal Phase HPLC and Corona Charged Aerosol Detection | Plante, M; Bailey, B; Acworth, I | PN71006, 2016 |
| Analysis of Emulsifiers in Foods by High Pressure Liquid Chromatography and Corona Charged Aerosol Detection | Plante, M; Bailey, B; Acworth, I; Crafts, C | PN70995, 2016 |
| Monitoring Peptide PEGylation by HPLC with Charged Aerosol Detection | Thomas, D; Acworth, I; Meier, S; Kaboord, B; Yang, H; Fisher, C | PN72093, 2016 |
| Application of Charged Aerosol HPLC Detection in Biopharmaceutical Analysis | Kopaciewicz, B; Thomas, D; Bailey, B: Zhang, Qi; Plante, M; Acworth, I | PN71803, 2015 |
| Analysis of Silicone Oils by High Performance Liquid Chromatography and Corona Charged Aerosol Detection | Plante, M; Bailey, B; Acworth, I | PN70538, 2013 |
| Analysis of Cationic Surfactants on the Acclaim Surfactant Plus Column | Foley, D; Faulkner, W | AN20574, 2012 |
| Acclaim™ Surfactant Column Product Manual | Thermo Scientific | Man065530, 2013 |
| Evaluation of Methods for the Characterization and Quantification of Polysorbates and Impurities Along with Other Surfactants and Emulsifiers Used in the Food and Pharmaceutical Industries | Plante, M; Fitchett, A; Hvizd, M | LPN2991, 2011 |

**Peer reviewed journals**

| Title | Authors | Publication |
|---|---|---|
| Development of a rapid and reliable analytical method for screening poloxamer 188 for use in cell culture process. | Bareford, L.; Peng, H.; Ali, A.; Kolwyck, D.; Dickens, J. | *Biotechnol. Prog.* **2019**, In Press. |
| Mechanism for the reduced dissolution of ritonavir tablets by sodium lauryl sulfate | Guo, Y.; Wang, C.; Dun, J.; Du, L.; Hawley, M.; Sun. C. C. | *J. Pharm. Sci.* **2019**, *108,* 516–524. |
| On-line coupling of hydrophobic interaction column with reverse phase column -charged aerosol detector/mass spectrometer to characterize polysorbates in therapeutic protein formulations | He, Y.; Brown, P.; Bailey Piatchek, M. R.; Carroll, J. A.; Jones, M. T. | *J. Chromatogr. A* **2019**, *1586,* 72–81. |
| Microwave-assisted synthesis and characterization of stearic acid sucrose ester: A bio-based surfactant | Kondamudi, N.; MsDougal, O. M. | *J. Surfact. Deterg.* **2019**, *22*, 721–729. |
| Factors influencing polysorbate's sensitivity against enzymatic hydrolysis and oxidative degradation | Kranz, W.; Wuchner, K.; Corradini, E.; Berger, M.; Hawe, A. | *J. Pharm. Sci.* **2019**, *108*, 2022–2032 |
| Quantitative structure-property relationship modeling of polar analytes lacking UV chromophores to charged aerosol detector response | Schilling, K.; Krmar, J.; Maljuric, N.; Pawellek, R.; Protic, A.; Holzgrabe, U. | *Anal. Bioanal. Chem.* **2019**, *411*, 2945–2959 |
| Multi-arm PEG-maleimide conjugation intermediate characterization and hydrolysis study by a selective HPLC method | Wang, J.; Yang, S.; Zhang, K. | *J. Pharm. Biomed. Anal. 164,* **2019**, 452–459. |
| Universal response quantification approach using a Corona Charged Aerosol Detector (CAD) - Application on linear and cyclic oligomers extractable from polycondensate plastics polyesters, polyamides and polyarylsulfones | Eckardt, M.; Kubicova, M.; Simat, T. J. | *J. Chromatogr. A* **2018**, *1572,* 187–202. |
| Simultaneous determination of anionic, amphoteric and cationic surfactants mixtures in surface water | Paun, I.; Iancu, V. I.; Cruceru, L.; Niculescu, M.; Chiriac, F. L. | *Ecolib.* **2018** |
| Influence of charged aerosol detector instrument settings on the ultra-high-performance liquid chromatography analysis of fatty acids in polysorbate 80 | Schilling, K., Pawellek, R., Lovejoy, K., Muellner, T., Holzgrabe, U. | *J. Chromatogr. A* **2018**, *1576,* 58–66. |
| PEG quantitation using reversed-phase High-Performance Liquid Chromatography and Charged Aerosol Detection | Smith, M. C.; Clogston, J. D. | *Meth. Mol. Biol.* **2018**, *1682,* 49–55. |
| Characterization of polydisperse macrogols and macrogol-based excipients via HPLC and charged aerosol detection | Theiss, C.; Holzgrabe, U. | *J. Pharm. Biomed. Anal.* **2018**, *160,* 212–221. |
| Textiles -- Method for the detection and determination of alkylphenol ethoxylates (APEO) -- Part 2: Method using NPLC | | *ISO 18254-2:2018* |

# Peer reviewed journal references



**Peer reviewed journals**

| Title | Authors | Publication |
|---|---|---|
| Pilot scale production of a phospholipid-enriched dairy ingredient by means of an optimised integrated process employing enzymatic hydrolysis, ultrafiltration and super-critical fluid extraction | Barry, K. M.; Dinan, T. G.; Kelly, P. M. | *Innov. Food Sci. Emerg. Tech.* **2017**, *41,* 301–306. |
| Applications of charged aerosol detection for characterization of industrial polymers | Cools, P.; Brooijmans, T. | *John Wiley & Sons, Inc.,* **2017** |
| Polymers and surfactants | Kou, D.; Manius, G.; Tian, H., Chokshi, H. P. | *John Wiley & Sons, Inc.,* **2017** |
| Impact of mono- and poly-ester fractions on polysorbate quantitation using mixed-mode HPLC-CAD/ELSD and the fluorescence micelle assay | Lippold, S.; Koshari, S. H. S.; Kopf, R.; Schuller, R.; Buckel, T.; Zarraga, I. E.; Koehn, H. | *J. Pharm. Biomed. Anal.* **2017**, *132,* 24–34. |
| Trends on analytical characterization of polysorbates and their degradation products in biopharmaceutical formulations | Martos, A.; Koch, W.; Jiskoot, W.; Wuchner, K.; Winter, G.; Friess, W.; Hawe, A. | *J. Pharm. Sci.* **2017**, *106,* 1722–1735 |
| High performance liquid chromatography-charged aerosol detection applying an inverse gradient for quantification of rhamnolipid biosurfactants | Behrens, B.; Baune, M.; Jungkeit, J.; Tiso, T.; Blank, L. M.; Hayen, H. | *J. Chromatogr. A* **2016**, *1455,* 125–132 |
| Residual host cell protein promotes polysorbate 20 degradation in a sulfatase drug product leading to free fatty acid particles | Dixit, N.; Salamat-Miller, N.; Salinas, P. A.; Taylor, K. D.; Basu, S. K. | *J. Pharm. Sci.* **2016**, *105,* 1657–1666. |
| Fatty acid composition analysis in polysorbate 80 with high performance liquid chromatography coupled to charged aerosol detection | Ilko, D.; Braun, A.; Germershaus, O.; Meinel, L.; Holzgrabe, U. | *Eur. J. Pharm. Biopharm.* **2015**, *94,* 569–574. |
| Simple and rapid high performance liquid chromatography method for the determination of polidocanol as bulk product and in pharmaceutical polymer matrices using charged aerosol detection | Ilko, D.; Puhl, S.; Meinel, L.; Germershaus, O.; Holzgrabe, U. | *J. Pharm. Biomed. Anal.* **2015**, *104,* 17–20. |
| Separation and quantification of phospholipid and neutral lipid classes by HPLC–CAD: Application to egg yolk lipids | Kielbowicz, G.; Trziszka, T.; Wawrzenczyk, C. | *J. Liq. Chromatogr.* **2015**, *38,* 898–903 |
| A highly sensitive method for the quantitation of polysorbate 20 and 80 to study the compatibility between polysorbates and m-cresol in the peptide formulation | Shi, S.; Chen, Z.; Rizzo, J. M.; Semple, A.; Mittal, S. | *J. Anal. Bioanal. Tech.* **2015**, *407,* 3705–3710. |
| Quantitative analysis of PEG-functionalized colloidal gold nanoparticles using charged aerosol detection | Smith, M. C.; Crist, R. M.; Clogston, J. D.; McNeil, S. E. | *Anal. Bioanal. Chem.* **2014**, *86, 5150–5157.* |
| Characterization and stability study of polysorbate 20 in therapeutic monoclonal antibody formulation by multidimensional ultrahigh-performance liquid chromatography-Charged Aerosol Detection-Mass Spectrometry | Li, Y.; Hewitt, D.; Lentz, Y. K.; Ji, J. Y. A.; Zhang, T. Y.; Zhang, K. | *Anal. Chem.* **2014**, *449,* 42–44. |

# Peer reviewed journal references



## Peer reviewed journals

| Title | Authors | Publication |
|---|---|---|
| Chromatographic methods for characterization of poly (ethylene glycol)-modified polyamidoamine dendrimers | Park, E. J.; Cho, H., Kim, S. W.; Na, D. H. | *Anal. Biochem.* **2014**, *449,* 42–44. |
| Simple and efficient profiling of phospholipids in phospholipase D-modified soy lecithin by HPLC with Charged Aerosol Detection | Kiełbowicz, G.; Micek, P.; Wawrzenczyk, C. | *J. Am. Oil Chem. Soc.* **2013**, *90,* 951–957. |
| A new liquid chromatography method with charge aerosol detector (CAD) for the determination of phospholipid classes. Application to milk phospholipids | Kiełbowicz, G.; Micek, P.; Wawrzenczyk, C. | *Talanta* **2013**, *105,* 28–33. |
| Analysis of ionic surfactants by HPLC with evaporative light scattering detection and charged aerosol detection | Kim, BH; Jang, JB; Moon, DC | *J. Liq. Chromatogr. Rel. Technol.* **2013**, *36,* 1000–1012. |
| Design of experiments and multivariate analysis for evaluation of reversed-phase high-performance liquid chromatography with charged aerosol detection of sucrose caprate regioisomers | Lie, A.; Wimmer, R.; Pedersen L. H. | *J. Chromatogr. A* **2013**, *1281,* 67–72. |
| An easy way to a fast universal method for surfactant analysis | Steiner, F., Plante, M., Bailey, B., Acworth, I. N. | *LCGC North America* [Online], **2012**, *8,* 2–9. |
| Investigating the stability of the nonionic surfactants tocopheryl polyethylene glycol succinate and sucrose laurate by HPLC-MS, DAD, and CAD | Christiansen, A.; Backensfeld, T.; Kuhn, S.; Weitschies, W. | *J. Pharm. Sci.* **2011**, *100,* 1773–1782. |
| Stability of the non-ionic surfactant polysorbate 80 investigated by HPLC-MS and charged aerosol detector | Christiansen, A.; Backensfeld, T.; Kühn, S.; Weitschies, W. | *Pharmazie* **2011**, *66,* 666–671. |
| Characterization of hydroxypropylmethylcellulose (HPMC) using comprehensive two-dimensional liquid chromatography | Greiderer, A.; Steeneken, L.; Aalbers, T.; Vivó-Truyols, G.; Schoenmakers, P. | *J. Chromatogr. A* **2011**, *1218,* 5787–5793. |
| Fast and sensitive determination of polysorbate 80 in solutions containing proteins | Fekete, S.; Ganzler, K.; Fekete, J. | *J. Pharm. Biomed. Anal.* **2010**, *52,* 672–679. |
| Analysis of polysorbate 80 using fast HPLC and charged aerosol detection | Lobback, C.; Backensfeild, T.; Funke, A.; Weltschies, W. | *J. Pharm. Biomed. Anal.* **2010**, *52,* 672–679. |
| Alternative sample-introduction technique to avoid breakthrough in gradient-elution liquid chromatography of polymers | Reingruber, E.; Bedani, F.; Buchberger, W.; Schoenmakers, P. | *J. Chromatogr. A* **2010**, *1217,* 6595–6598. |
| Size exclusion chromatography with Corona charged aerosol detector for the analysis of polyethylene glycol polymer | Kou, D.; Manius, G.; Zhan, S.; Chokshi, H. P. | *J. Chromatogr. A* **2009**, *1216,* 5424–5428. |

**Peer reviewed journals**

| Title | Authors | Publication |
|---|---|---|
| Quantification of pegylated phospholipids decorating polymeric microcapsules of perfluorooctyl bromide by reverse phase HPLC with a charged aerosol detector | Diaz-Lopez, R.; Libong, D.; Tsapis, N.; Fattal, E.; Chaminade, P. | *J. Pharm. Biomed. Anal.* **2008**, *48,* 702–707. |
| Obstacles and pitfalls in the PEGylation of therapeutic proteins | Gaberc-Porekar, V.; Zore, I.; Podobnik, B.; Menart, V. | *Curr. Opin. Drug Discovery Dev.* **2008**, *11,* 242–250. |
| PEGylation of cholecystokinin prolongs its anorectic effect in rats | León-Tamariz, F.; Verbaeys, I.; Van Boven, M.; De Cuyper, M.; Buyse, J.; Clynen, E.; Cokelaere, M. | *Peptides* **2007**, *28,* 1003–1011. |
| Quantitative determination of nonionic surfactants with CAD | Lobback, C.; Backensfeld, T.; Funke, A.; Weitschies, W. | *Chromatogr. Tech.* **2007**, *10,* 18–20. |

Find out more at **www.thermofisher.com/CAD**

**For Research Use Only. Not for use in diagnostic procedures.** ©2019 Thermo Fisher Scientific Inc. All rights reserved. Zonyl FSO Fluorosurfactant and related products are trademarked and owned by DuPont. Triton is a registered trademark of Union Carbide Co., Inc. Corexit is a registered trademark of Nalco Holding Company. Kolliphor and Pluronic are registered trademarks of BASF Corp. Synperonic and Tween are registered trademarks of Croda International PLC. Alkest and Canarcel are registered trademarks of Oxiteno. Variquat is a registered trademark of Evonik Industries AG. All other trademarks are the property of Thermo Fisher Scientific and its subsidiaries unless otherwise specified. This information is presented as an example of the capabilities of Thermo Fisher Scientific Inc. products. It is not intended to encourage use of these products in any manners that might infringe the intellectual property rights of others. Specifications, terms and pricing are subject to change. Not all products are available in all countries. Please consult your local sales representative for details. **AN71101-E1019S**

ThermoFisher SCIENTIFIC