

Use of Reversed Phase and HILIC in the 2D-LC Separation of Cationic, Anionic, and Hydrotropic Surfactants

Introduction

Surfactants are frequently found in pharmaceutical and biopharmaceutical drug applications as well as in common household products. Because they can be polar, non-polar, or amphoteric, the structural diversity of the surfactants and complexity of the sample matrix can make their separation and identification by HPLC challenging. Historically, this has been achieved using Reversed Phase Chromatography supplemented with Ion Exchange Chromatography. When performed individually, these techniques can be used to characterize the surfactant profile. This Application Note illustrates the effectiveness of a reversed phase high throughput column, TSKgel® ODS-140HTP, in series with a TSKgel NH₂-100 HILIC column, for the separation of polar and non-polar surfactants in a single injection using 2D-LC.

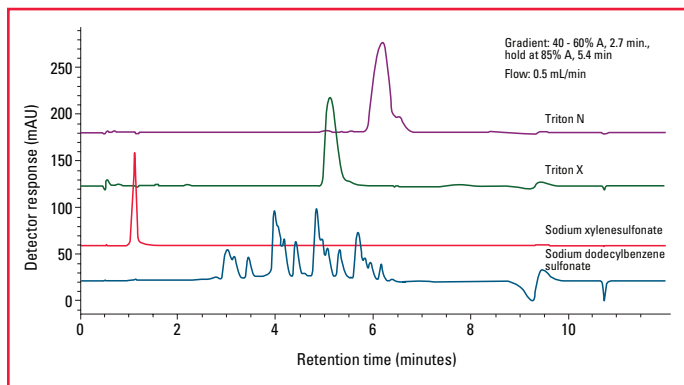
Materials and Methods

Columns:	TSKgel ODS-140HTP, 2.3 μm, 2.1 mm ID × 5 cm TSKgel NH ₂ -100, 3 μm, 2 mm ID × 15 cm
Instrument:	Agilent 1200 HPLC system run by Chemstation (ver B.04.02)
Mobile phase:	A: CH ₃ CN B: 100 mmol/L ammonium acetate, pH 5.4
Gradient:	40-60% A, 2.7 minutes, hold at 85% A, 5.4 minutes
Flow rate:	0.5 mL/min
Detection:	UV @ 280 nm, 254 nm, and 210 nm FLD λ _{ex} 280 nm, λ _{em} 350 nm
Temperature:	30 °C
Injection vol.:	1 μL
Samples:	Triton™ X Triton N sodium xylenesulfonate sodium dodecylbenzene sulfonate

Results and Discussion

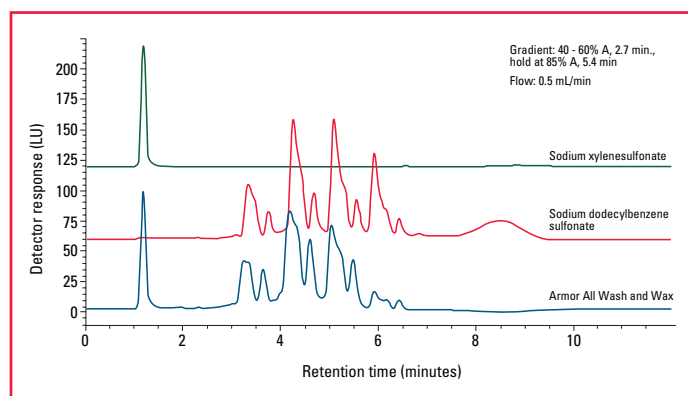
Figure 1 depicts the separation of four surfactants using the TSKgel ODS-140HTP and TSKgel NH₂-100 columns in series. The use of a shallow, non-linear gradient of 40-60% CH₃CN in 2.7 minutes, followed by an isocratic hold at 85% CH₃CN till 5.4 minutes, allowed for high resolution of the sodium dodecylbenzene sulfonate impurities and good retention of sodium xylene sulfonate as well. This figure illustrates the utility of 2D-LC methodology for separating a diverse mixture of surfactants.

Figure 1. 2D-LC Separation of Surfactants using the TSKgel ODS-140HTP and the TSKgel NH₂-100 Columns



Due to the success in resolving the surfactant standards in a single run as explained above, the usefulness of this technique in detecting the corresponding UV-absorbing surfactant ingredients in an actual commercial household product was evaluated. **Figure 2** illustrates the characterization of the surfactant profile of Armor All™ Wash and Wax using the 2D-LC methodology described above. As shown, the use of the TSKgel ODS-140HTP and TSKgel NH₂-100 columns in series yielded excellent separation and retention of the anionic surfactant sodium dodecylbenzene sulfonate and the hydrotropic surfactant sodium xylene sulfonate present in the Armor All formulation. Additionally, the use of fluorescence detection (λ_{ex}: 225 nm, λ_{em}: 300-400 nm) allowed for increased sensitivity of the low level surfactants found in the product.

Figure 2. Characterization of surfactant profile in Armor All Wash and Wax using 2D-LC with the TSKgel ODS-140HTP and TSKgel NH₂-100 columns



Conclusions

The purpose of this study was to illustrate the effectiveness of 2D RP/HILIC for the separation of anionic, cationic, and hydrotropic surfactants. The use of the TSKgel ODS-140HTP column in series with the TSKgel NH₂-100 column allows for the simultaneous retention and separation of both polar and non-polar hydrotropic, nonionic, and anionic surfactants in a single injection. The characterization of surfactants in a common household product is also successfully performed with the TSKgel ODS-140HTP and TSKgel NH₂-100 columns, allowing for strong retention and good peak shape of various surfactants with no interference from other matrix compounds.

Tosoh Bioscience and TSKgel are registered trademarks of Tosoh Corporation.
The Armor All trademark is a registered trademark of The Armor All/STP Products Company



TOSOH BIOSCIENCE

TOSOH

TOSOH BIOSCIENCE LLC
3604 Horizon Drive, Suite 100
King of Prussia, PA 19406
Tel: 800-366-4875
email: info.tbl@tosoh.com
www.tosohbioscience.com

AN70
0814