Sensitivity
Flexibility
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A New Generation of Evaporative Light-Scattering Detectors for Liquid Chromatography: Universality, Reliability and Cost-Effectiveness in Food Analysis - An Application Review in HPLC and U-HPLC

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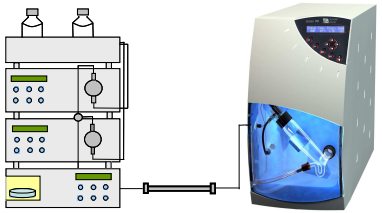
Abstract

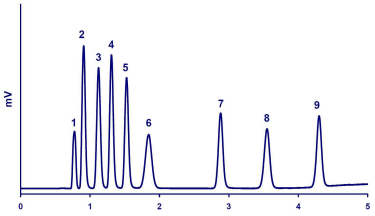
Food and beverage safety and compositional information during the full process requires increasing quantities of complete, reliable, fast and cost-effective analyses. Therefore, development of analytical methods using HPLC and U-HPLC gained an increasing importance in Food industry during the last decades. The goals of such analytical work are various, but are mainly oriented towards monitoring of the chemical and biochemical content in R&D, in quality control at all stages of production and preservation, and products authenticity.
Among the detectors available in Liquid Chromatography (LC), Evaporative Light-Scattering Detector (ELSD) became in recent years a well established instrument thanks to several theoretical studies based on fundamental investigations and numerous applications provided during the last thirty years. Indeed, ELSD is considered as a nearly Universal, powerful, reliable and cost-effective technique, and is ideally appropriate in Food industry for a great variety of LC applications containing chromophoric and non-chromophoric compounds. Today, the power of this detection mode is further extended with the ultimate model which proposes a genuine and efficient Low-Temperature technology (LT-ELSD™) combined with an innovative detection chamber, thus providing the highest sensitivities with all compounds including semi-volatile and thermo-labile ones.

To show the strength and the versatility of this ELSD model, several LC methods applicable to Food analysis are developed in this work. These applications use the most recent LC media, such as HILIC, sub-two-micron or fused-core particle phases, allowing outstanding separations and simultaneous analyses of a wide range of chemical and biochemical compounds. This work proposes several selected LC-ELSD methods and includes the analyses of the following groups of compounds:
- Sugars and polyols.
- Sweeteners.
- Lipids.
- Amino acids.
- Vitamins.
- Minerals and inorganic salts.
- Aroma chemical compounds; this last group emphasizes an unexpected detection mode for semi-volatile molecules characterized by high vapor pressures.



HPLC / LT-ELSD System

I - Sugars and Polyols

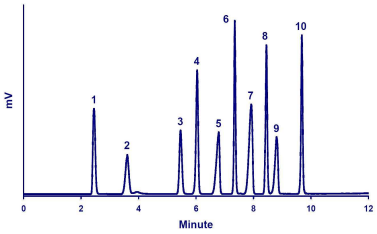


Chromatogram of the Fast and Simultaneous HPLC/LT-ELSD Analysis of Polyols, Mono- and Disaccharides.

Standard mixture: 1-Glycerol, 2-Rhamnose, 3-Arabinose, 4-Fructose, 5-Sorbitol, 6-Glucose, 7-Inositol, 8-Saccharose, 9-Maltose (500ppm each)
Injection volume: 2µL
Column: Imtakt UK-Amino (3µm, 2.0 x 150mm), 60°C
Flowrate: 0.8mL/min
Eluent: A: H2O - B: ACN
Gradient: 0-2 minutes: 10%A, 2-3 minutes: from 10%A to 20%A, 3-5 minutes: 20%A

Detector: SEDEX 90LT, 30°C, 3.5bar

II - Sweeteners: Natural and Artificial

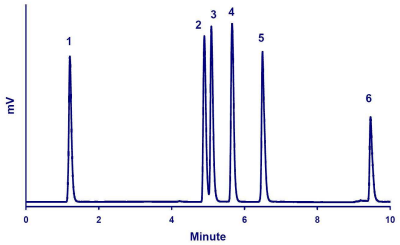


Chromatogram of the Simultaneous HILIC/LT-ELSD Analysis of Natural and Artificial Sweeteners including Polyols.

Standard mixture: 1-Erythritol, 2-Sucralose, 3-Sorbitol, 4-Neotame, 5-Acesulfame, 6-Neohesperidin dihydrochalcone, 7-Saccharin, 8-Aspartame, 9-Cyclamate, 10-Rebaudioside A (from *Stevia*) (500ppm each)
Injection volume: 2µL
Column: Hypersil GOLD HILIC (1.9µm, 2.1 x 150mm), 40°C
Flowrate: 0.5mL/min
Eluent: A: Ammonium acetate 50mM, pH5 - B: ACN
Gradient: 0-0.5 minute: 5%A, 0.5-10 minutes: from 5%A to 30%A

Detector: SEDEX 90LT, 35°C, 3.5bar

III - Lipids: Analytical Monitoring of Biodiesel Production

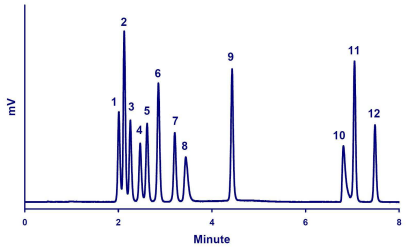


Chromatogram of the Normal-Phase HPLC/LT-ELSD Analysis of Tripalmitin Hydrolysis Products.

Standard mixture: 1-Tripalmitin, 2-1,3 Dipalmitin, 3 -1,2 Dipalmitin, 4-Palmitic acid, 5-Monopalmitin, 6-Glycerol (250ppm for 1, 2, 3, 4, 5, and 1000ppm for semi-volatile glycerol)
Injection volume: 2µL
Column: Syncronis Amino (1,7µm, 2,1 x 100mm), 30°C
Flowrate: 0.5mL/min
Eluent: A: Hexane - B: Acetone + 0.2% formic acid
Gradient: 0-1 minute 100%A, 1-6 minutes: from 100%A to 50%A, 6-8 minutes: from 50%A to 0%A

Detector: SEDEX 90LT, 28°C, 3.5bar

IV - Underivatized Amino Acids: Essential and Semi-Essential

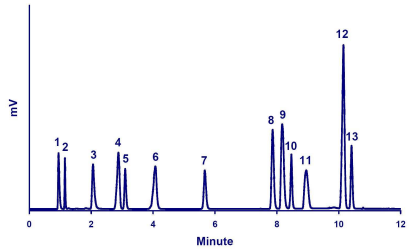


Chromatogram of the Direct HILIC/LT-ELSD Analysis of Essential and Semi-Essential Amino Acids.

Standard mixture: 1-PHE, 2-TRY, 3-LEU, 4-ILE, 5-MET, 6-TYR, 7-VAL, 8-CYS, 9-THR, 10-HIS, 11-ARG, 12-LYS (250ppm each)
Injection volume: 2µL
Column: Syncronis HILIC (1,7µm, 2,1 x 100mm), 30°C
Flowrate: 0.5mL/min
Eluent: A: Ammonium formate 100mM, pH3 - B: ACN
Gradient: 0-1 minute: 16%A, 1-2.5 minutes from 16%A to 25%A, 2.5-3.5 minutes: 25%A, 3.5-4 minutes: from 25%A to 30%A, 4-8 minutes: 30%A

Detector: SEDEX 90LT, 40°C, 3.5bar

V - Vitamins, Minerals, Sweeteners, Amino acids, Salts, Caffeine: Global Method for Food Additives and Food Supplements

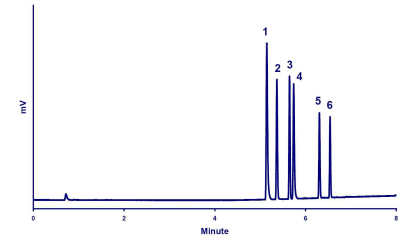


Chromatogram of the Simultaneous HILIC/LT-ELSD Analysis of Several Food Additives and Supplements.

Standard mixture: 1-Caffeine, 2-Vitamin B3, 3-Vitamin B6, 4-Chloride, 5-Erythritol, 6-Vitamin C, 7-Xylitol, 8-Potassium, 9-Sodium, 10-Rebaudioside A (from *Stevia*), 11-Taurine, 12-Magnesium, 13-Glutamic acid, (500ppm to 1000ppm each)
Injection volume: 1µL
Column: Ascentis Express HILIC (2.7µm, 2.1 x 150mm), 30°C
Flowrate: 0.5mL/min
Eluent: A: Ammonium formate 200mM, pH:3 - B: ACN
Gradient: 0-1 minute: 8%A, 1-8 minutes: from 8%A to 25%A, 8-12 minutes: 25%A

Detector: SEDEX 90LT, 40°C, 3.5bar

VI – Aroma Chemical Compounds



Chromatogram of the Simultaneous HPLC/LT-ELSD Analysis of Several Aroma Chemical Compounds.

Standard mixture: 1-Vanillic acid, 2-p-Hydroxybenzaldehyde, 3-Vanillin, 4-Syringaldehyde, 5-Ethylvanillin, 6-Coumarin
Injection volume: 1µL
Column: Halo C18 (2.7µm, 2.1 x 150mm), 25°C
Flowrate: 0.4mL/min
Eluent: A: H2O + 0.2% formic acid - B: ACN + 0.2% formic acid
Gradient: 0-1 minute: 5%B, 1-7 minutes: from 5%B to 90%B

Detector: SEDEX 90LT, 28°C, 3.5bar

Conclusion

. The examples developed in this work demonstrate the Universal applicability of LT-ELSD in HPLC for various types of both chromophoric and non-chromophoric compounds such as mono- and disaccharides, polyols, artificial and natural sweeteners, fatty acids, mono-, di- and triglycerides, amino acids, vitamins, minerals, inorganic salts, aroma chemicals and phenolic compounds and methylxanthine alkaloids. These applications using just a single Universal detector are straightforward to operate and do not require any additional equipment nor specific tedious sample preparation.
. The New SEDEX 90LT benefits from both the Low-Temperature technology and a new optical head design based on a high-performance laser, which results in an outstanding sensitivity increase with LODs typically down to the very low-nanogram levels and RSD in the range of 1-2%.
. Moreover, SEDEX 90LT allows the use of acetone which is less toxic and cheaper than many other organic eluents (such as acetonitrile), and which possesses excellent physical and chromatography properties.
. As a conclusion, the association of the most recent chromatography media and the new generation of high-performance ELSDs provides the analysts with a quite relevant and cost-effective solution to their separation and quantification challenges in Food area.