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Step-Down Gradient Improves the Chromatographic Separation of Sucrose Monocaprate Regioisomers

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Introduction
Sugar fatty acid esters have a broad range of industrial applications and are used in the food, cosmetic and pharmaceutical industries. These compounds can be synthesized by conventional chemical processes or by enzymatic methods in organic solvents. Their physical and chemical properties depend on fatty acid chain length and both position and degree of esterification. The sugar monoesters are the most important types of these compounds because of higher solubility in water compared to the corresponding oligoesters.

In analysing sugar fatty acid monoester syntheses, it is necessary to achieve separation of the regioisomers. Ritthitham et al. (2009) previously demonstrated that when using reversed-phase high-pressure liquid chromatography (RP-HPLC), a step down in the concentration of acetonitrile in the eluent, below the initial concentration, improved the separation of sucrose fatty acid monoesters significantly. They also provided the elution order for seven regioisomers of sucrose monocaprate. Sucrose, shown in Figure 1, has eight hydroxyl substituents, and can form up to eight monoester regioisomers.

Aim
The aim of the present study was to improve the chromatographic separation of sucrose monocaprate regioisomers using reversed-phase high-pressure liquid chromatography.

Method
A commercial sample of sucrose monocaprate (>95 %) was analysed on an HPLC-system (HP Series 1100) with evaporative light scattering detection (Alltech ELSD 800). The stationary phase was a C18 column (Waters Symmetry, 5 µm, 4.6x250mm) and gradients of acetonitrile (CH3CN) in water (H2O) were used as mobile phase.

Results
In the present investigation of sucrose monocaprate regioisomers analysed using RP-HPLC, a systematic study of the separation effects was performed. Three elution factors important for separation were identified:

A) Initial concentration of CH3CN in the eluent mixture
B) Duration of the initial isocratic elution
C) Step-down concentration of CH3CN in the eluent mixture

In one experiment elution factor C was varied while all other parameters were kept constant, while the effects of variation of elution factors A and B was investigated separately.

Conclusions
A step-down gradient can significantly improve the separation of sucrose monocaprate regioisomers in reversed-phase high-pressure liquid chromatography analysis.

The improvement achieved in the separation of sucrose monocaprate regioisomers in terms of retention times was in the area of 400 %.

References