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Application Note SI-01663

Removal of Salts prior to Carbohydrate Analysis

A Ewen

Pilot Laboratory, Test and Research Institute Pirmasens (PFI), Germany

Introduction

For enzymatic hydrolysis of celluloses and hemicelluloses to occur, it is necessary to utilize buffer solutions (eg Sorensen's buffer). However, to perform HPLC analysis on the resulting monosaccharides, the samples must first be desalinated to protect the column. These are loaded with cations (eg Pb2+, Ca2+, Na+, K+) to achieve a certain selectivity. To permanently ensure a troublefree analysis and constant separation, it is necessary to remove all salts from the samples to prevent the exchange and rinse out of the loaded cations. By processing the samples using StratoSpheres™ PL-MIXED MP cartridges, which contain strong cation exchange and strong anion exchange functionality, it is possible to remove all buffer ingredients with a high recovery of the carbohydrates.

Figure 1 shows the concentration of phosphorus (as representative for the salts) in the different eluted fractions (each 1.0 mL). Note that there is no breakthrough of salts when PL-MIXED MP with 500 mg bed mass is used.

Figure 2 shows the concentration of carbohydrates in the different eluted fractions (each 1.0 mL). Table 1 shows their recovery results following elution with 5.0 mL of deionized water, and subsequent HPLC-RID analysis.

In summary, the StratoSpheres PL-MIXED MP SPE strong anion and cation exchanger allows good retention of the buffer ingredients resulting in a high recovery of the sugars.

Conditions

Cartridge: StratoSpheres PL-MIXED MP SPE (pn: PL3540-C610)

Dimensions: 500 mg / 6.0 mL

Capacity: 1.2 mmol (0.6 mmol cations and 0.6 mmol anions)

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Conditioning: 1 column volume deionized water

Sample Application: 1.0 mL solution of sugars (cellobiose, glucose,

xylose, galactose, arabinose) in Sorensen's buffer (corresponds to 0.1 mmol anions and 0.1 mmol

cations)

Concentrations: Sugars: 1 g/L each

Sorensen's Buffer: 4.54 g/L $\rm KH_2PO_4$ and 11.9 g/L

Na₂HPO₄•12H₂O

Elution/Rinsing Out: 5.0 mL of deionized water

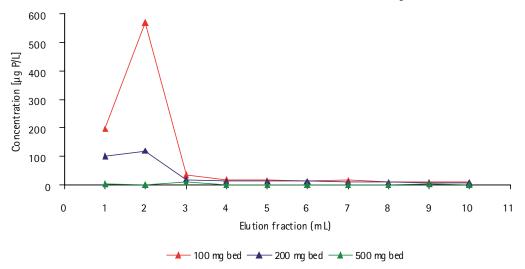


Figure 1. Concentration of phosphorus (ICP-OES analysis) in the different eluted fractions (each 1.0 mL). No breakthrough of salts when 500 mg PL-MIXED MP is used.

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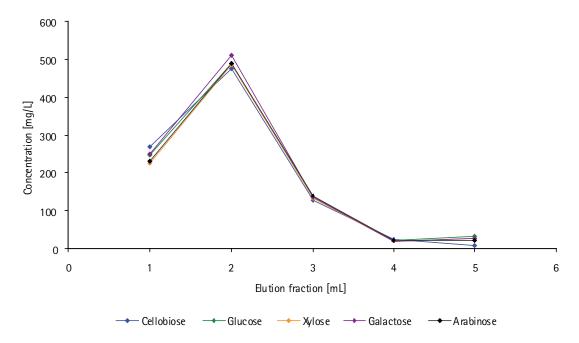


Figure 2. Concentration of carbohydrates in the different eluted fractions (each 1.0 mL). The chart shows clearly that the carbohydrates are nearly completely eluted (respectively rinsed out) with 5.0 mL water.

Table 1. Recovery results of the carbohydrates after elution / rinsing out with 5.0 mL water (HPLC-RI analysis).

Carbohydrate	Recovery (%)
Cellobiose	90
Glucose	93
Xylose	89
Galactose	94
Arabinose	91

These data represent typical results.
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