

Application Note

Introduction

Carbohydrates are commonly known as sugars and widely used in food preparation. They fall into several catagories, monosaccharides, such as Glucose, Fructose and Galactose, and more complex disaccharides, such as Sucrose, Maltose and Lactose., and then longer chain oligosaccharides.

Monosaccharides are called "simple sugars", if 2 monosaccharides form a glycosidic bond with the loss of a water molecule then these are classified as disaccharides. This linkage may continue to grow to produce oligosaccharides of great length.

Figure 1. Sucrose, a disaccharide of glucose (left) and fructose (right)

Sugars have been associated with many health factors and risks, most notibly, blood glucose levels (Diabetics), immune system suppression, obesity and cardiovascular disease. Therefore their analysis is of great interest in clinical applications as well as in food analysis.

Experimental

Sugars are difficult to detect due to their lack of chromophore, therefore refractive index detectors are typically employed. Elevated temperatures allow the rapid elution of the sugars.

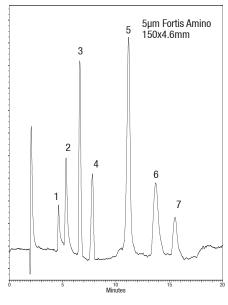
Analytical Method:

p/n FNH-050705 Mobile phase 75 : 25 ACN : Water Flow Rate: 1.0ml/min

Temp: 30°C Detection: RI

Column: 5µm Fortis™ Amino 150 x 4.6 mm

Carbohydrates

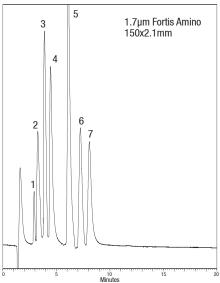


UHPLC Run:

Column: 1.7µm Fortis™ Amino 150 x 2.1 mm

p/n FNH-020701 Mobile phase 75 : 25 ACN : Water Flow Rate: 0.3ml/min

Temp: 40°C Detection: RI



In the UHPLC method resolution of the sugars is high and sensitivity of the analysis is also

improved over the analytical method. Pressure remains low as the combination of high organic and raised temperature allow the use of the longer small particle column.

Compounds

- 1. Ribose
- 2. Xylose
- 3. Fructose
- 4. Glucose
- 5. Sucrose
- 6. Maltose
- 7. Lactose

Results

7 monosaccharide and disaccharide sugars were analysed on the Fortis Amino column in a simple mobile phase system using RI as the detection method.

This was then transferred to a 1.7um Fortis Amino column to show the ability to speed up the analysis in UHPLC mode, whilst gaining good sensitivity and resolution. Retention of the 7 sugars reduced from 16minutes to less than 9minutes just with a simple mobile phase velocity change. Shorter column length or even higher flow rate could help speed this separation even further.

Conclusion

Fortis[™] Amino columns are ideal for the analysis of sugars in simple mobile phases. The amino bonding is robust, reproducible and stable even at the elevated pressures required in UHPLC, upto 18,000 psi.

Many food samples can be analysed for carbohydrate content once the resolution of the main monosaccharides and disaccharides is optimised.

Clinical laboratories will find use of the Fortis Amino as an L8 column specifically for Pharmacopia sugar methods, such as the analysis of Lactulose.