Thermo Scientific Dionex CarboPac PA1 Columns for Mono-, and Di- and Oligosaccharide Analysis

HPLC Columns for Quantitative Analysis of Mono- and Di- and Oligosaccharides Over an Extensive Range of Sample Matrices

- Detection by pulsed amperometric detection (PAD); no derivatization needed
- Exceptionally high sensitivity
- Approved for use in a variety of official methods for the analysis of foods (AOAC Methods 995.13, 996.04, 997.08, 2000.11, 2000.17 and 2001.02)



Unique Column Chemistry for Optimal Performance

The Thermo Scientific™ Dionex™

CarboPac™ PA1 Column is a specialized anion-exchange columns designed to be used with PAD to deliver high resolution separations of di- and some oligosaccharides. The resins consist of 10 µm diameter nonporous beads covered with a fine latex of functionalized Thermo Scientific™ Dionex™ MicroBead™ resin. This pellicular resin structure permits excellent mass transfer, resulting in high resolution chromatography and rapid reequilibration.

The Dionex CarboPac PA1 column is a rugged all-purpose column suitable for determining monosaccharides and disaccharides in a variety of matrices. It is also the column of choice for high resolution separations of linear polysaccharides. The Dionex CarboPac PA1 column is available in microbore, standard bore and semi-preparative formats.



The Dionex CarboPac PA1 (2 mm) does an excellent job for the analysis of common sugars in various fruit juice samples as demonstrated in Figures 1A and B using pulsed amperometric detection (HPAE-PAD). Note that each juice samples was diluted a thousand fold with deionized water to avoid any overloading of the column and the detector.

Glycoprotein Monosaccharide Compositional Analysis

The Dionex CarboPac PA1 column can be used for the quantification of amino, acidic, and neutral monosaccharides, especially those derived from glycoconjugates. (See Figure 2).

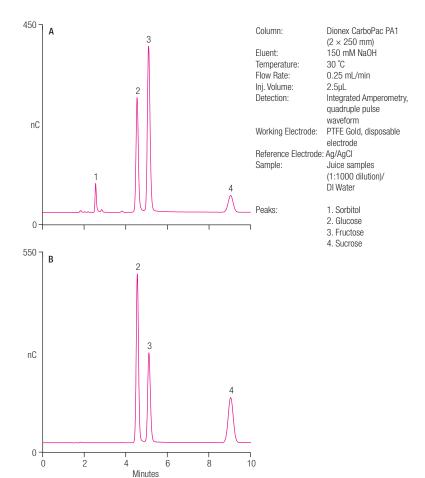


Figure 1. Separation of sugars in A) apple juice and B) cranberry juice using the Dionex CarboPac PA1 (2 mm) column.

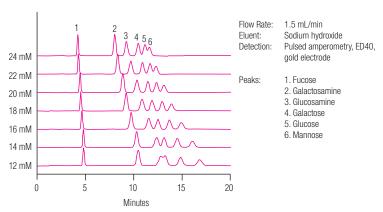


Figure 2. Dionex CarboPac PA1 column for glycoprotein monosaccharides.

Linear Polysaccharides

Chain length distribution is an important parameter in starch characterization. HPAE-PAD technology featuring the Dionex CarboPac PA1 column delivers single-residue resolution of linear polysaccharides up to a degree of polymerization (DP) of at least 60. Figure 3 shows the chain length distribution of a sample of purified inulin using the Dionex CarboPac PA1 column. Commercial inulin products have DPs that have been tailored for a particular end use; therefore, it is important to determine the chain length distribution during product development and production and for quality control of the end product. Figure 3 illustrates the excellent resolution that can be acheived for DP values up to 50 or more for purified inulin.

Acidic Oligosaccharides

Sialic acids comprise a large family of N- and *O*- substituted neuraminic acids. They occupy terminal positions on many mammalian glycoproteins and glycolipid oligosaccharides. When a glycoprotein loses sialic acid residues, it has a reduced serum half-life and in some cases reduced activity. Therefore it is important to know the sialic acid content of a glycoprotein when assaying its function or its efficacy as a pharmaceutical therapeutic. HPAE-PAD is an effective way to determine Neu5Ac and Neu5Gc without derivatization, and can be performed with either the Dionex CarboPac PA1 or Dionex CarboPac PA10 column.

The elution of acidic sugars from the Dionex CarboPac PA1 column requires stronger eluents than those used for neutral sugars. This is usually accomplished by the addition of sodium acetate to the sodium hydroxide eluent. See Figure 4. Sodium acetate accelerates the elution of strongly bound species without interfering with PAD.

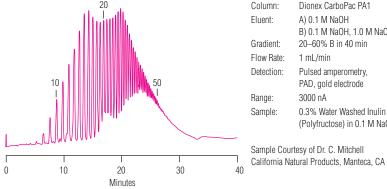
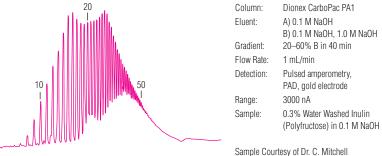


Figure 3. Chromatographic analysis of purified inulin.



Column: Dionex CarboPac PA1 Peaks: 1. α -D-Galactosamine -1-P 1.13 µg Flow Rate: 1.0 mL/min 2. α -D-Glucosamine-1-P 0.45 α-D-Galactose-1-P 1.75 Detector: Pulsed amperometry, lpha-D-Glucose-1-P 1.75 PAD, gold electrode α-D-Ribose-1-P 1.46 Range: 10.000 nA R- D-Glucose-1-P 1.75 Eluent A: 100 mM NaOH D-Glucosamine-6-p 3 75 100 mM NaOH, Eluent B: D-Galactose-6-P 2.04 1.0 M NaOAc D-Glucose-6-P 1.25 Gradient: D-Fructose-1-P 0.96 %A D-Fructose-6-P 0.42 11 lpha-D-Glucuronic acid-1-P 3.08 0 90 12 10 13. α-D-Glucose-1,6-Di P 1.06 20 80 20 ß-D-Fructose-2,6-Di P 0.92 12 30 50 50 15. D-Fructose-1,6-Di P 3 30 10 20 40

Figure 4. Analysis of mono- and diphosphorylated monosaccharides on the Dionex CarboPac PA1 column.

Minutes

Official Methods Using the Dionex CarboPac PA1 Column

The Dionex CarboPac PA1 column has been approved for use in a number of official methods, including ISO/DIS 11292 for coffee authenticity, International Committee of Uniform Methods of Sugar Analysis (ICUMSA) for sugars in molasses, and AOAC Methods 996.04 and 2000.11. Figure 5 shows an example of an isocratic method to determine sugars in molasses. Table 1 lists official food methods using Thermo Scientific technology.

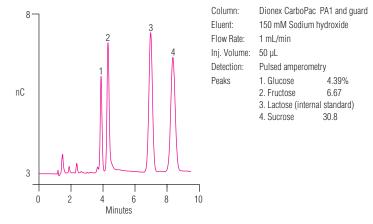


Figure 5. Isocratic separation of sugars found in sugarcane molasses. Official method of the International Commission for Uniform Methods of Sugar Analysis.

Table 1. Official food methods using Thermo Scientific technology.

Analysis	Thermo Scientific Technology	Official Method
Sugars in molasses	HPAE-PAD/Dionex CarboPac PA1	AOAC Method 996.04 ICUMSA (1994)
Carbohydrates in soluble coffee	HPAE-PAD/ Dionex CarboPac PA1	AOAC Method 995.13 ISO 11292
Fructans in food	HPAE-PAD/ Dionex CarboPac PA1	AOAC Method 997.08
Anions in beer by IC	IC with suppressed conductivity	Analytica-EBC International Method
Polydextrose	HPAE-PAD/ Dionex CarboPac PA1	AOAC Method 2000.11
Transgalacto-oligosaccharides	HPAE-PAD/ Dionex CarboPac PA1	AOAC method validation study in progress

SPECIFICATIONS		
Dionex CarboPac PA1 Colum	n	
Resin Composition:	10 μm diameter substrate (polystyrene 2% crosslinked with divinylbenzene) agglomerated with 500-nm MicroBead quaternary ammonium functionalized latex (5% crosslinked)	
Anion Exchange Capacity:	Approximately 100 μ eq/column (4 \times 250 mm analytical column)	
Maximum Operating Pressure:	4000 psi (27.9 MPa)	
Chemical Compatibility:	pH 0-14, up to 2% of common HPLC solvents	

Ordering Information

In the U.S., call (800) 346-6390 or contact the Thermo Fisher Scientific Regional Office nearest you. Outside the U.S., order through your local Thermo Fisher Scientific office or distributor. Refer to the following part numbers. Thermo Fisher Scientific can also make special order Dionex CarboPac columns to your specifications; call for more information.

Description	Part Number	
Dionex CarboPac PA1 Analytical Columns		
Analytical Column (4 × 250 mm)	035391	
Guard Column (4 × 50 mm)	043096	
Analytical Column (2 × 250 mm)	057178	
Guard Column (2 × 50 mm)	057179	
Dionex CarboPac PA1 Semi-Preparative Column (9 × 250 mm)	039686	
Dionex CarboPac PA1 Semi-Preparative Column (22 × 250 mm)	SP2866	
Dionex CarboPac PA1 Guard (9 × 50 mm)	063501	
Dionex AminoTrap Column (4 × 50 mm)	046122	
Dionex BorateTrap Column (4 × 50 mm)	047078	
MonoSaccharide Standard		
Thermo Scientific™ Dionex™ MonoStandards™ Mixture of Six, 100 nmol each. Contains fucose, galactosamine HCl, glucosamine HCl, galactose, glucose and mannose	043162	

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