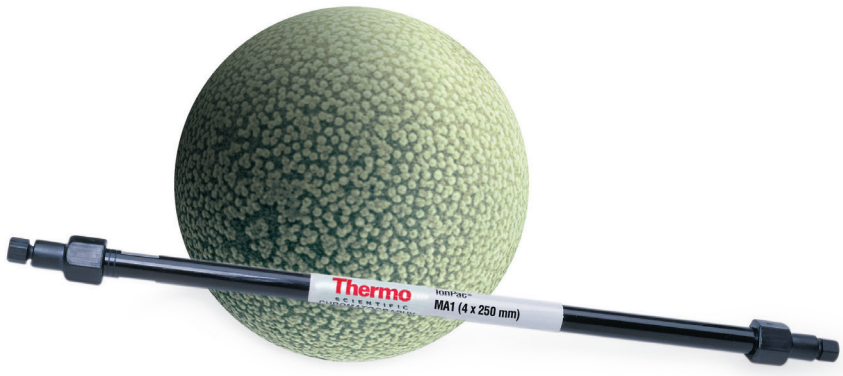
Thermo Scientific Dionex CarboPac MA1 Column

HPLC columns for the analysis of weakly ionizable carbohydrates.

- Simple methods with pulsed amperometric detection
- Reduced mono- and disaccharides in food products and physiological samples
- Unique selectivity
- Higher sensitivity than refractive index detection
- No sample derivatization required
- Ambient temperature



Simple Methods with Pulsed Amperometric Detection

The Thermo Scienfitic™ Dionex™ CarboPac™ MA1 column is specifically designed to separate reduced monosaccharides and disaccharides commonly found in food products, as well as those found in physiological samples. Using a simple hydroxide eluent, analytes can be separated isocratically or with gradient elution. The low flow rate ensures low eluent consumption and reduces errors associated with eluent preparation.

The Dionex CarboPac MA1 column is also well suited to the separation of reduced glycoconjugate samples after ß-elimination. Designed to retain and separate reduced carbohydrates, the Dionex CarboPac MA1 column separates sugar alcohols as their anions by high performance anion-exchange chromatography (HPAE). Detection of these complex compounds has been optimized with the use of pulsed amperometric detection (PAD).

Thermo SCIENTIFIC

Reduced Mono- and Disaccharides in Food Products

Several alditols, including sorbitol and mannitol, occur naturally in foods. These alditols can be separated isocratically and detected using pulsed amperometry (see Figure 1). Mannose, glucose, and galactose were added to the sample to demonstrate that aldoses exhibit greater retention than their reduced alditol forms. Gradient elution can be used, allowing fast run times of selected analytes.

Commercial Sweeteners

Isomaltitol and glucopyranosylmannitol are found in commercial sweeteners. In Figure 2, lactitol is centered between isomaltitol and glucopyranosylmannitol. Quantification of lactitol and isomaltitol is improved and total run time is reduced by a step change to 600 mM sodium hydroxide.

Sucralose

Sucralose, developed jointly by Tate and Lyle and McNeil Specialty Products, is a high-intensity sweetener with 400–800 times the sweetness of sucrose. Sucralose is manufactured by selective chlorination of sucrose; it is currently the only non-nutritive sweetener based on sucrose. The product was approved in Canada in September 1991 for use in a variety of food products. Figure 3 shows an analysis of trace impurities in a Sucralose sample using HPAE-PAD.

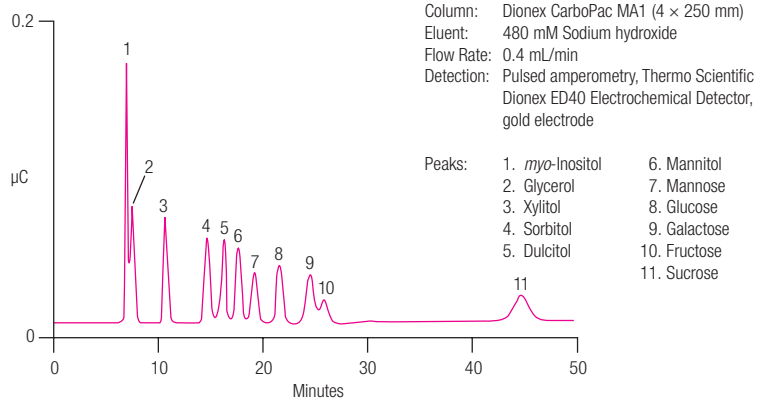


Figure 1. Isocratic separation and pulsed amperometric detection of common, naturally occurring food alditols and aldoses. Mannose, glucose, and galactose were added to the sample to demonstrate that aldoses exhibit greater retention than their reduced alditol forms.

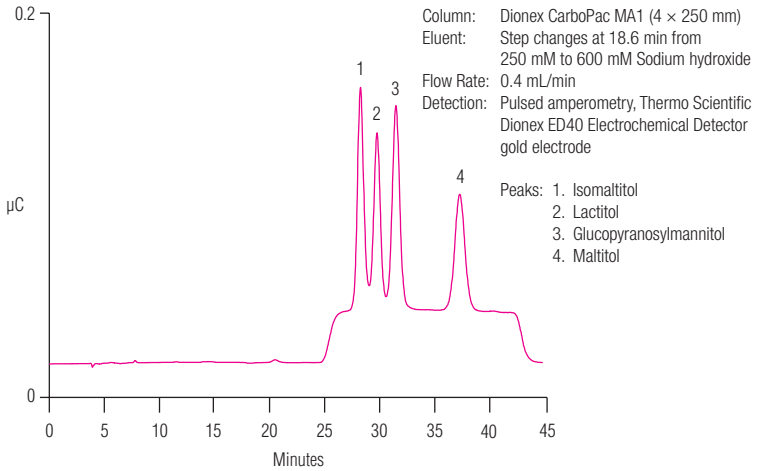


Figure 2. Separation of compounds found in commercial sweeteners.

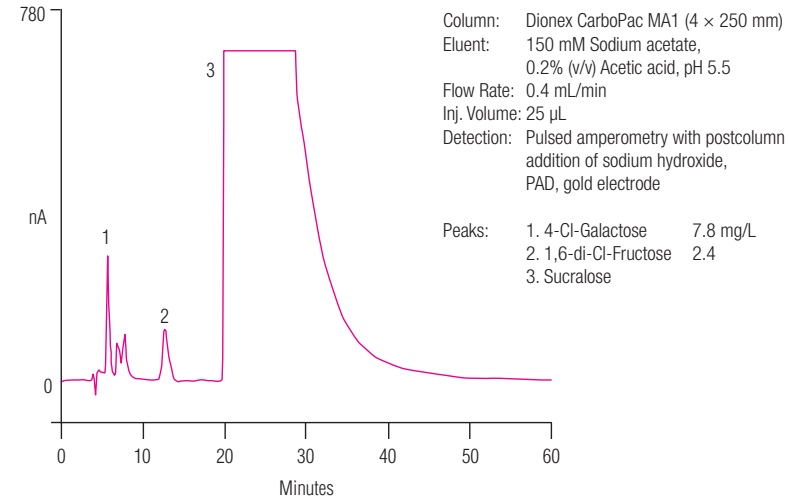


Figure 3. Analysis of sucralose.

Vanillin

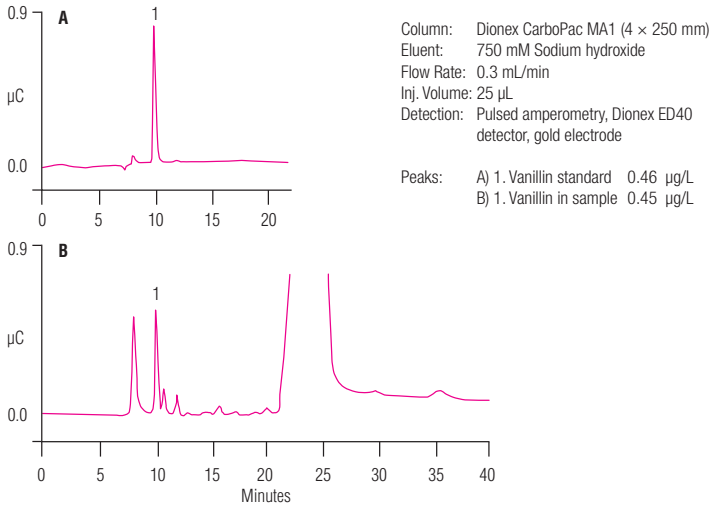
Vanillin is the most widely used flavoring in food products. Adulteration of natural vanillin by synthetic products is a major concern. Vanillin is an aromatic aldehyde and can be detected by pulsed amperometric detection with high sensitivity and specificity. This is particularly advantageous when determinations are made in complex food matrices since clean-up procedures can be minimized. Figure 4 shows the determination of vanillin in a sample of yogurt. Sample preparation involved a 100-fold weight/volume dilution and filtration (0.45 µg/L).

Sugar Alcohols in Candy

Sugar alcohols are often used as sweeteners in dietetic confectionery products for diabetics. They are also non-cariogenic (i.e., they do not cause tooth decay) since they are not fermented by bacteria in the mouth. However, the use of sugar alcohols in foods is regulated because they sometimes exhibit laxative or diuretic properties. As shown in Figure 5, sorbitol and mannitol are easily determined, interference-free, in a hard candy (boiled sweets) sample after dissolution and dilution.

Carbohydrates in Chewing Gum

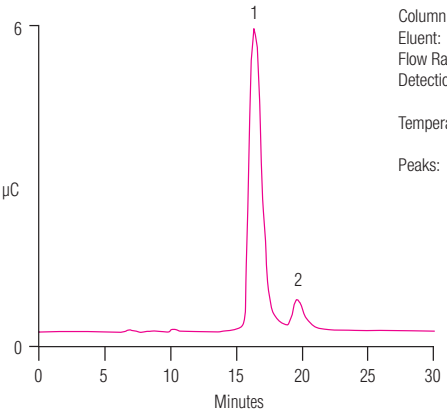
Figure 6 shows an analysis of chewing gum. Glycerol, sorbitol, mannitol, and glucose were determined directly in the chewing gum sample after sonication with deionized water, followed by passage through a Thermo Scientific™ Dionex™ OnGuard™ A Cartridge cartridge and filtration through a 0.45 µm filter.



Column: Dionex CarboPac MA1 (4 × 250 mm)
Eluent: 750 mM Sodium hydroxide
Flow Rate: 0.3 mL/min
Inj. Volume: 25 µL
Detection: Pulsed amperometry, Dionex ED40 detector, gold electrode

Peaks: A) 1. Vanillin standard 0.46 µg/L
B) 1. Vanillin in sample 0.45 µg/L

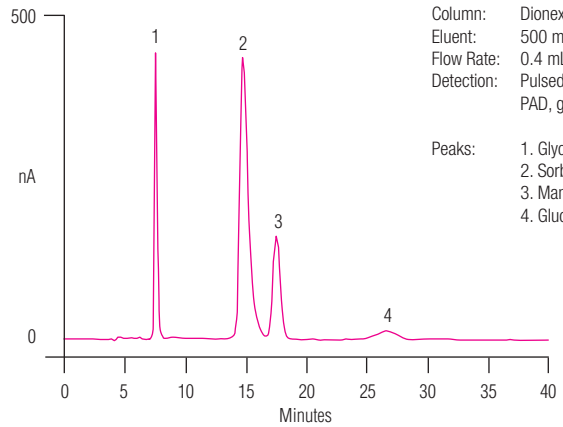
Figure 4. Determination of vanillin in yogurt.



Column: Dionex CarboPac MA1 (4 × 250 mm)
Eluent: 500 mM Sodium hydroxide
Flow Rate: 0.4 mL/min
Detection: Pulsed amperometry, Dionex ED40 detector, gold electrode
Temperature: Ambient

Peaks: 1. Sorbitol 106 µg/L
2. Mannitol 17

Figure 5. Determination of sugar alcohols in sugarless hard candy.



Column: Dionex CarboPac MA1 (4 × 250 mm)
Eluent: 500 mM Sodium hydroxide
Flow Rate: 0.4 mL/min
Detection: Pulsed amperometry, PAD, gold electrode

Peaks: 1. Glycerol 0.39 µg/L
2. Sorbitol 1.1
3. Mannitol 0.34
4. Glucose 0.06

Figure 6. Determination of carbohydrates in chewing gum.

Sugars in Fruit Juices

The Dionex CarboPac MA1 column is specifically tailored for the determination of sugar alcohols, but mono- and disaccharides can also be determined in the same run. Sorbitol is present in high levels in apple juice relative to other fruit juices. It is a useful marker for the detection of adulteration of more costly juices with apple juice. The only sample preparation required is dilution and filtration (Figure 7).

Reduced Mono- and Disaccharides in Physiological Fluids

Inositol derivatives are ubiquitous in living tissues and physiological fluids. The Dionex CarboPac MA1 column separates *myo*-inositol from *scyllo*-inositol under isocratic conditions. Separation of inositols from other alditols such as erythritol, arabitol, and sorbitol can be achieved under isocratic conditions, but sample throughput can be enhanced by gradient elution (Figure 8).

Glycoconjugates

The carbohydrate moieties of glycoproteins and glycolipids can be analyzed by HPAE-PAD. Figure 9 illustrates the identification of monosaccharides derived from glycoprotein oligosaccharides. Oligosaccharides are released by reductive ß-elimination, which converts only the terminal peptide-linked sugar to the alditol form. The terminally reduced oligosaccharide is chemically or enzymatically digested into a mixture containing one alditol and one or more non-reduced monosaccharides.

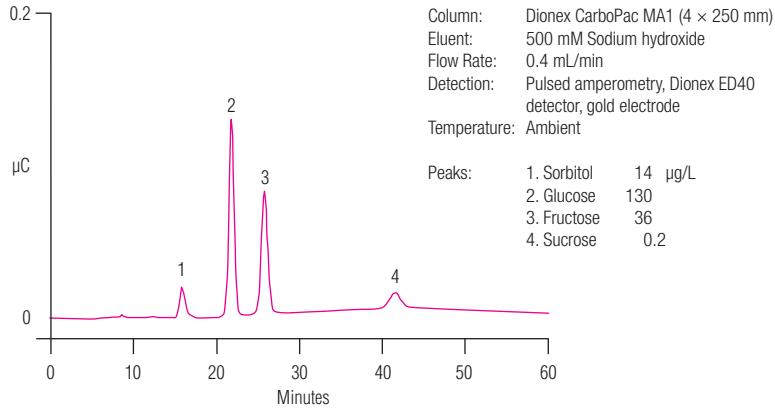


Figure 7. Determination of sugars and sorbitol in fruit juice.

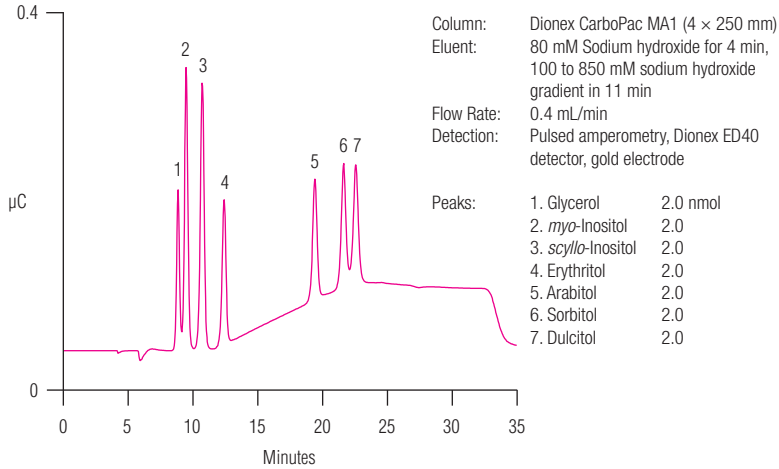


Figure 8. Separation of alditols typically found in living tissues and physiological fluids.

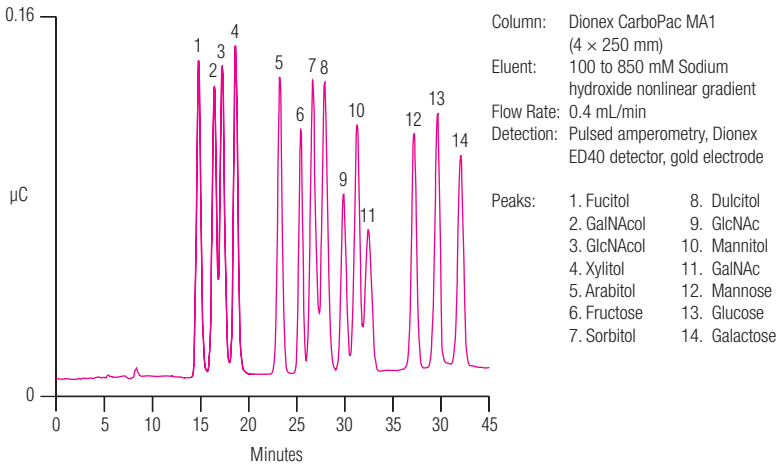


Figure 9. Gradient separation of alditols and aldoses potentially released by hydrolysis of ß-eliminated oligosaccharides. Arabitol serves as an internal standard.

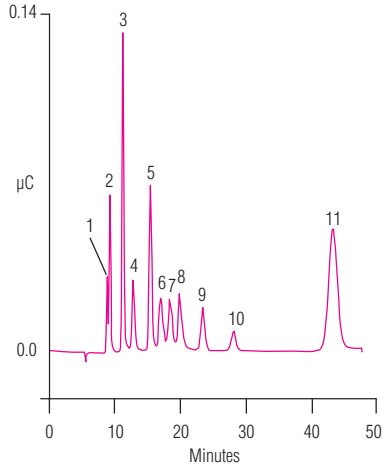
No Sample Derivatization Required

The use of sugar alcohols (also known as polyols) as alternative sweeteners is increasing rapidly for dietetic foods and products such as chewing gum because of their non-cariogenic properties. Nutritional labeling requirements for sugar alcohols are presently optional in the U.S., but as with sugars, total sugar alcohol content is listed. Therefore, minor sugar alcohols have to be determined (see Figure 10). GC methods for sugar alcohols have been developed but are complicated by the requirement for derivatization. The only official method in existence is AOAC 973.28, a GC method for sorbitol.

A simpler, direct approach that does not require derivatization is shown in Figure 10. The column was designed to allow sugar alcohols to be eluted before sugars; therefore sugar alcohols and sugars can be determined in the same run. Sorbitol, mannitol, and xylitol are monosaccharide polyols in widespread use. Erythritol is expected to receive regulatory approval as a commercial sweetener in the near future. It has all the advantages of other polyols with the added benefit that only 10% or less is metabolized, giving it only 10% of the caloric value of sucrose.

Rugged, Reliable Analyses with Guaranteed Performance

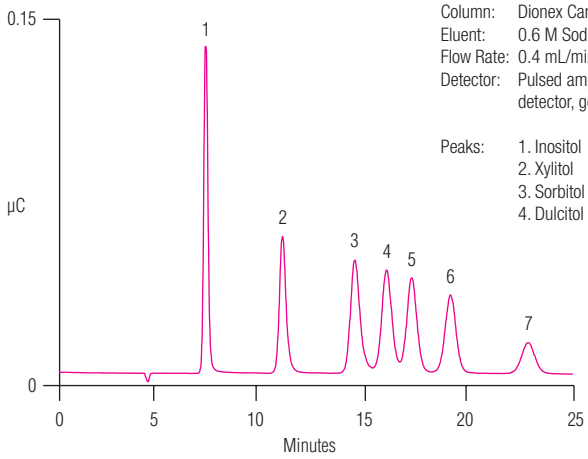
The entire manufacturing process (including resin synthesis, amination, and packing of the chromatography columns) is carefully controlled to ensure that every Dionex CarboPac MA1 column delivers reproducible performance. The Dionex CarboPac MA1 columns are tested with a representative set of alditols and aldoses (Figure 11) to ensure lot-to-lot consistency. Even with challenging samples, the Dionex CarboPac MA1 columns delivers run-to-run reproducibility and a long useful lifetime (Figure 12).



Column: Dionex CarboPac MA1 (4 × 250 mm)
Eluent: 500 mM Sodium hydroxide
Flow Rate: 0.4 mL/min
Inj. Volume: 10 µL
Detection: Pulsed amperometry, Dionex ED40 detector, gold electrode

Peaks:
1. *myo*-Inositol
2. Glycerol
3. *i*-Erythritol
4. Xylitol
5. Arabitol
6. Sorbitol
7. Dulcitol
8. Mannitol
9. Glucose
10. Fructose
11. Sucrose

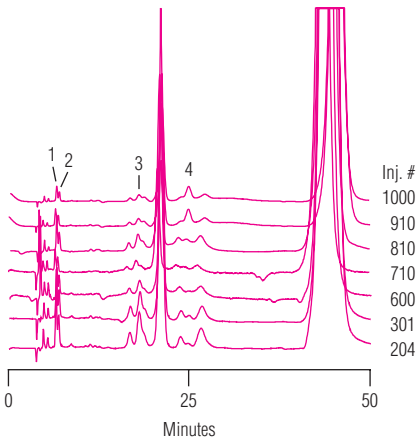
Figure 10. HPAE separation of sugars and sugar alcohols without the need for derivatization.



Column: Dionex CarboPac MA1 (4 × 250 mm)
Eluent: 0.6 M Sodium hydroxide
Flow Rate: 0.4 mL/min
Detector: Pulsed amperometry, Dionex ED40 detector, gold electrode

Peaks:
1. Inositol
2. Xylitol
3. Sorbitol
4. Dulcitol
5. Mannitol
6. Glucose
7. Fructose

Figure 11. Isocratic separation of a representative set of alditols and aldoses ensures lot-to-lot reproducibility in the manufacturing of the Dionex CarboPac MA1 column.



Column: Dionex CarboPac MA1 (4 × 250 mm)
Eluent: 480 mM Sodium hydroxide
Flow Rate: 0.4 mL/min
Detector: Pulsed amperometry, Dionex ED40 detector, gold electrode

Peaks:
1. Inositol
2. Glycerol
3. Mannitol
4. Fructose

Figure 12. Separation of carbohydrates from instant coffee. In this run-to-run reproducibility study, an instant coffee sample was injected repeatedly onto the Dionex CarboPac MA1 column. Retention time RSDs for all components were less than 0.8% over a one month period using multiple eluent preparations.

Easy Isocratic Operation

Retention of carbohydrates on the Dionex CarboPac MA1 column can be varied with eluent concentration (see Table 1). Note that the elution order for several sugars changes with the sodium hydroxide concentration. Table 1 is a guide for the development of separations of specific sets of analytes.

Table 1. K' Values of selected analytes.

	Eluent Concentration (M NaOH)				
Analyte	**0.05**	**0.14**	**0.25**	**0.38**	**0.50**
Glycerol	1.13	0.99	0.89	0.80	0.72
myo-Inositol	1.32	1.08	0.86	0.69	0.56
scyllo-Inositol	1.63	1.30	1.02	0.81	0.64
GlcNol	1.81	1.40	1.09	0.89	0.75
Fucitol	1.94	1.63	1.40	1.18	1.05
Erythritol	2.02	1.71	1.44	1.25	1.13
GalNol	2.29	1.81	1.39	1.13	0.95
GalNAcol	2.35	1.81	1.38	1.12	0.95
GlcNAcol	2.61	1.96	1.48	1.18	0.95
Xylitol	3.09	2.48	1.95	1.59	1.35
Arabitol	4.69	3.62	2.76	2.24	1.92
Sorbitol	6.43	4.72	3.33	2.55	2.06
Dulcitol	6.52	5.04	3.73	2.87	2.26
Adonitol	7.09	5.31	3.83	2.96	2.43
Mannitol	8.98	6.38	4.37	3.28	2.63
Fucose	10.34	4.72	2.52	1.69	1.25
Isomaltitol	12.22	8.15	4.89	3.30	2.43
Lactitol	14.97	9.61	5.49	3.57	2.43
gp-Mannitol	15.66	10.36	6.18	4.15	3.05
GalN	18.56	7.16	3.39	2.13	1.48
GlcN	20.88	7.71	3.61	2.24	1.55
Maltitol	31.21	17.25	8.80	5.44	3.67
Glucose	> 32	15.70	7.19	4.31	2.91
Mannose	> 32	13.55	6.15	3.72	2.53
Galactose	> 32	17.82	8.25	4.99	3.43

Product Specifications

SPECIFICATIONS

Resin Composition
7.5 µm diameter vinylbenzyl chloride/divinylbenzene macroporous substrate fully functionalized with an alkyl quaternary ammonium group (15% crosslinked)

Anion-Exchange Capacity
1450 µeq/column (4 × 250 mm analytical column)

Maximum Operating Pressure
2000 psi (14 MPa)

Chemical Compatibility
pH 0–14; incompatible with solvents and anionic detergents

Temperature Range
4–50 °C

Test Procedure
Baseline resolution of inositol, xylitol, sorbitol, dulcitol, mannitol, glucose, and fructose

Typical Operating Conditions
1400 psi at 0.4 mL/min (guard and analytical columns)

Recommended Operating Temperature
Ambient

Recommended Flow Rate:
0.2–0.5 mL/min

Eluents
Acetate or hydroxide only

Ordering Information

For more information or to place an order, contact the Thermo Scientific Dionex Products office nearest you or your local distributor. Phone numbers and addresses for worldwide subsidiaries can be found in the About Us section of www.thermoscientific.com.

Analytical, Capillary, and Guard Columns	Part Number
Dionex CarboPac MA1 Analytical Column (4 × 250 mm)	044066
Dionex CarboPac MA1 Guard Column (4 × 50 mm)	044067

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Africa +43 1 333 50 34 0
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Austria +43 810 282 206
Belgium +32 53 73 42 41
Brazil +55 11 3731 5140
Canada +1 800 530 8447
China 800 810 5118 (free call domestic)
400 650 5118
Denmark +45 70 23 62 60
Europe-Other +43 1 333 50 34 0
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