Carbohydrate Analysis

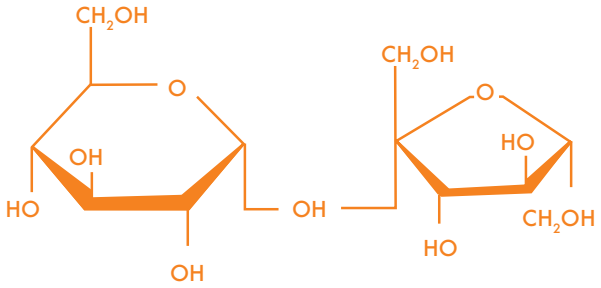
Prevail™ Carbohydrate ES HPLC Columns and ELSD

Brochure #467A

www.discoverysciences.com

GRACE

The Challenge of Separating and Detecting Carbohydrates



Silica-based amino columns are powerful sugar separators, but they degrade under conventional aqueous mobile phase conditions required for sugar analyses. The result is loss of retention, poor peak shape, and column voiding. The problem is made more apparent when using ELS and MS detectors because these detectors respond to non-volatile bonded phase loss as baseline noise and/or baseline shift.

Because they lack chromophores, carbohydrates present a particular detection challenge. The traditional approaches to carbohydrate detection are either differential refractive index (RI) or low-wavelength UV (190 – 205nm). The poor sensitivity and gradient incompatibility of RI detection make it less than ideal for carbohydrate analysis by HPLC. Low-wavelength UV also lacks sensitivity and places stringent requirements on the optical properties of the mobile phase. Both techniques are subject to severe baseline shifts and long equilibration times as the detector responds to spectroscopic changes in the solvent.

Benefits of Evaporative Light Scattering Detection (ELSD) for Carbohydrate Analysis

- **More sensitive than RI and low-wavelength UV**
- **Gradient compatible**
- **Stable baselines, even with gradients**
- **Solvent choice not limited by UV-cutoff**

The Evaporative Light Scattering Detector (ELSD) is a universal HPLC detector that detects any compound less volatile than the mobile phase to low nanogram levels. It has many advantages over RI and low-wavelength UV detection for carbohydrate analyses.

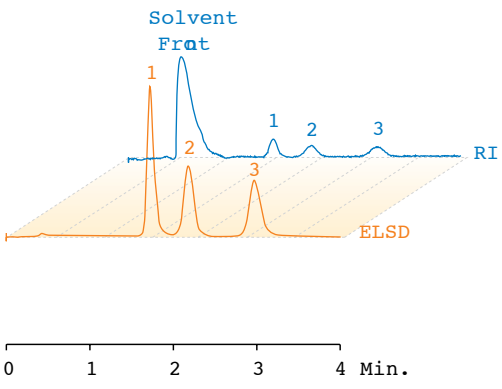
Since carbohydrates are not good UV absorbers, they must be monitored at low wavelengths. This can be hindered by the mobile phase solvent's UV cutoff and poor baseline stability. With ELSD, the mobile phase evaporates before detection, so it is blind to spectroscopic properties and the compositional changes in the mobile phase. The result is quiet, stable baselines, and better sensitivity than with low-wavelength UV detection.

Although refractive index (RI) detection is universal in nature, its lack of sensitivity and gradient incompatibility make it less than ideal for carbohydrate analysis. Evaporation of the mobile phase before detection makes the ELSD gradient compatible for improved resolution and faster separations. In addition, ELSDs can be 10 to 1000 times more sensitive than RI detectors.

The ELSD Improves Baseline Stability and Detection Sensitivity Compared to RI and Low-Wavelength UV For Carbohydrate Analyses

CHROM 10278
CHROM 10277

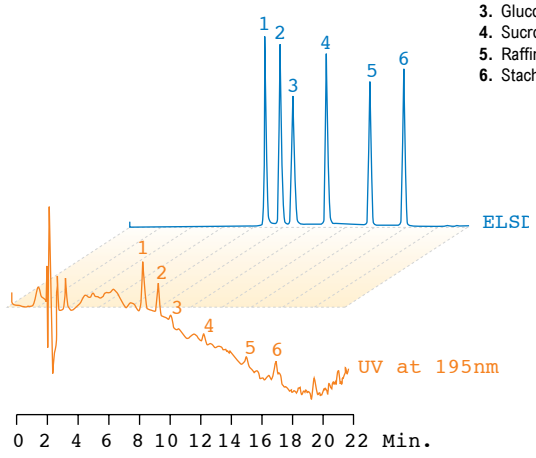
1. Fructose
2. Glucose
3. Sucrose



Column:	Prevail™ Carbohydrate ES, 5µm, 53 x 7.0mm Rocket™ (Part No. **35104**)
Mobile Phase:	Acetonitrile:Water (75:25)
Flow Rate:	2.0mL/min
Column Temp:	30°C

CHROM 9355
CHROM 9380

1. Mannitol
2. Fructose
3. Glucose
4. Sucrose
5. Raffinose
6. Stachyose



Column:	Prevail™ Carbohydrate ES, 250 x 4.6mm
Mobile Phase:	**A:** Acetonitrile **B:** Water
Gradient:	**Time:** 0 20 25 / **%B:** 20 50 50
Flow Rate:	1.0mL/min
Column Temp:	Ambient

Benefits of Prevail™ Carbohydrate ES Columns for Carbohydrate Analysis

- **Highly stable and rugged**
- **Stable baselines, even with gradients**
- **Outstanding resolution and peak shapes**

The Alltech® Prevail™ Carbohydrate ES Column is free from the problems that plague other carbohydrate columns. It is extremely rugged and maintains stable baselines with all detectors, including the ELSD. The polymeric Prevail™ Column shows none of the chemical degradation that limits the lifetime of silica-based amino columns.

Quiet Gradient Baselines

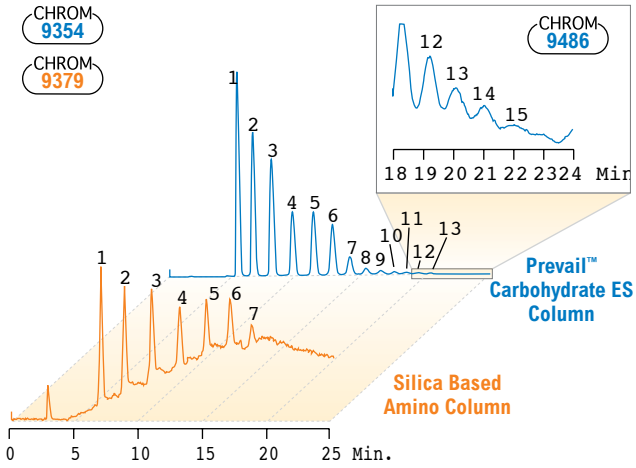
The Prevail™ Carbohydrate ES Column works very well for isocratic separations, especially when used with the ELSD. Gradients with Prevail™ Carbohydrate ES Columns show flat, quiet baselines, free from noise and drift. Not only are gradients possible, they're recommended because they offer powerful selectivity, reduced run times, and enhanced detector sensitivity as peak efficiencies are maximized. Finally, solvents may be chosen for their chromatography advantages instead of their spectral properties.

Exceptional Resolution and Peak Shape

Prevail™ Carbohydrate ES Columns are versatile enough for mono- and oligosaccharides and sugar alcohols. They quickly resolve fructose, glucose, lactose, maltose, and oligosaccharides with high peak efficiencies. Further, Prevail™ Carbohydrate ES Columns produce single peaks for reducing sugars like glucose at ambient temperatures, eliminating the need for column heating.

Enhance Detector Sensitivity With Stable Gradient Baselines

CHROM 9354
CHROM 9379



1. Dextrose
2. Maltose
3. Maltotriose
4. Maltotetraose
5. Maltopentaose
6. Maltohexaose
7. Maltoheptaose
8. Maltooctaose
9. Maltononaose
10. Maltodecaose
11. Maltoundecaose
12. Maltododecaose
13. Maltotridecaose
14. Maltotetradecaose
15. Maltopentadecaose

Column: 250 x 4.6mm as identified
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	15
%B:	35	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Reduce Run Times Using Alternative Solvents

CHROM 9350

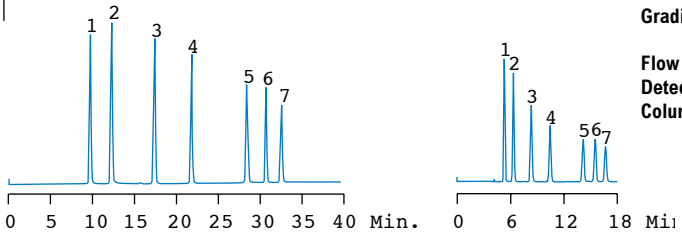
Unmodified Mobile Phase

Column: Prevail™ Carbohydrate ES, 5µm 250 x 4.6mm (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	50
%B:	20	35

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

1. Fructose
2. Glucose
3. Sucrose
4. Maltose
5. Maltotriose
6. Maltotetraose
7. Maltopentaose



CHROM 9360

Modified Mobile Phase with Acetone

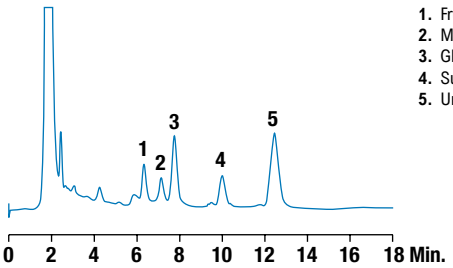
Column: Prevail™ Carbohydrate ES, 5µm 250 x 4.6mm (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile:Acetone (75:25) **B:** Water
Gradient:

Time:	0	18
%B:	24	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Conquistador® Tequila

CHROM 10585

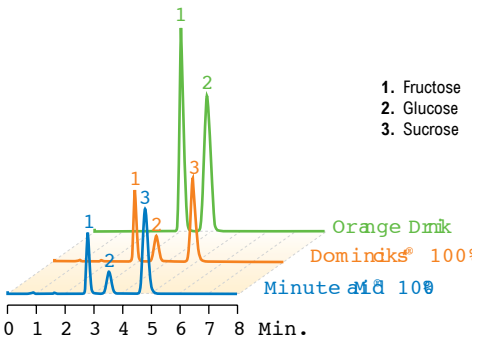


1. Fructose
2. Mannitol
3. Glucose
4. Sucrose
5. Unknown

Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm (Part No. **35101**)
Mobile Phase: Acetonitrile : Water (75:25)
Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Orange Juice Profiles

CHROM 9532
CHROM 9531
CHROM 9530

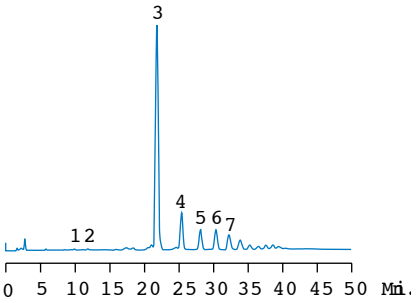


1. Fructose
2. Glucose
3. Sucrose

Column: Prevail™ Carbohydrate ES, 5µm, 53 x 7.0mm Rocket™ (Part No. **35104**)
Mobile Phase: Acetonitrile:Water (75:25)
Flow Rate: 1.3mL/min
Detector: ELSD
Column Temp: Ambient

Beer

CHROM 9351



1. Fructose
2. Glucose
3. Maltotetraose
4. Maltotriose
5. Maltopentaose
6. Maltohexaose
7. Maltoheptaose

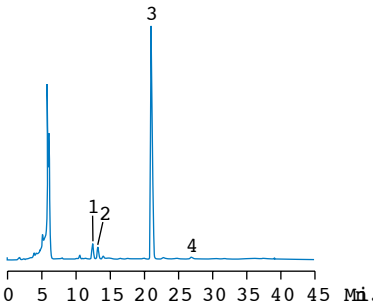
Column: Prevail™ Carbohydrate ES, 5µm 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	50
%B:	20	35

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Wine - Concord Grape

CHROM 9476



1. Sorbitol
2. Fructose
3. Maltose
4. Inositol

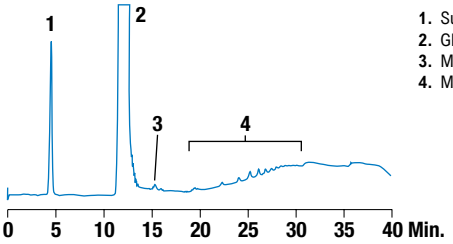
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:**Water
Gradient:

Time:	0	60
%B:	17	35

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Splenda® Sweetener

CHROM 10531



1. Sucralose
2. Glucose
3. Maltose
4. Maltodextrins

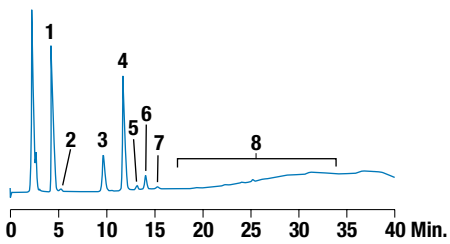
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	5	10	15	25
%B:	20	20	30	30	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Pepsi One® Cola

CHROM 10530



1. Sucralose
2. Unknown 1
3. Fructose
4. Glucose
5. Unknown 2
6. Sucrose
7. Maltose
8. Maltodextrins

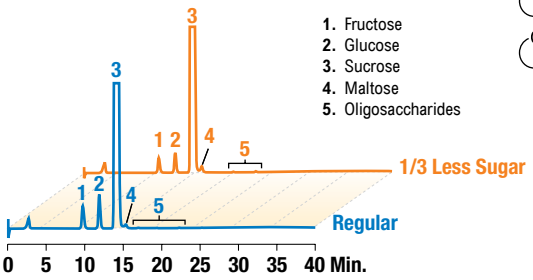
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	5	10	15	25
%B:	20	20	30	30	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Kellogg's Frosted Flakes® Cereal Regular vs. 1/3 less sugar

CHROM 10528
CHROM 10527



1. Fructose
2. Glucose
3. Sucrose
4. Maltose
5. Oligosaccharides

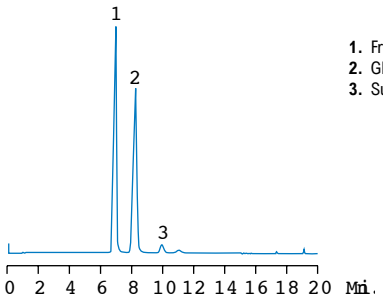
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	5	10	15	25
%B:	20	20	30	30	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Clover Honey

CHROM 9365



1. Fructose
2. Glucose
3. Sucrose

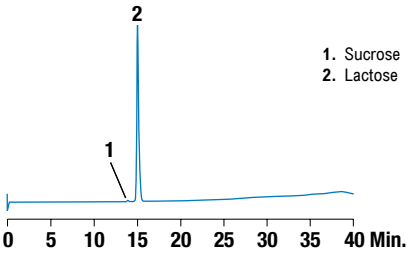
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	15
%B:	25	40

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Infant Formula

CHROM 10529



1. Sucrose
2. Lactose

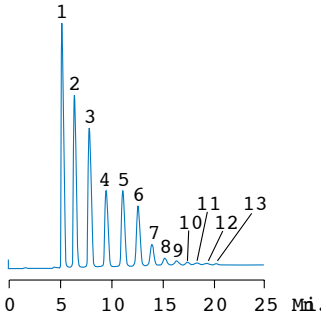
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	5	10	15	25
%B:	20	20	30	30	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Corn Syrup Solids

CHROM 9354



1. Dextrose
2. Maltose
3. Maltotriose
4. Maltotetraose
5. Maltopentaose
6. Maltohexaose
7. Maltoheptaose
8. Maltooctaose
9. Maltononaose
10. Maltodecaose
11. Maltoundecaose
12. Maltododecaose
13. Maltotridecaose

Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

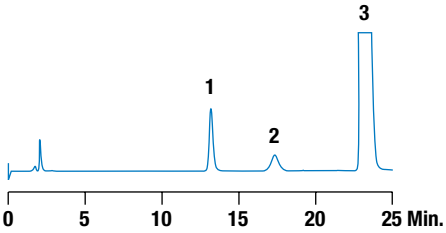
Time:	0	15
%B:	35	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Flavor Sample Solution

Chromatogram courtesy of Charles Stewart, Firmenich Inc.

CHROM 10579



1. Fructose
2. Glucose
3. Sucrose

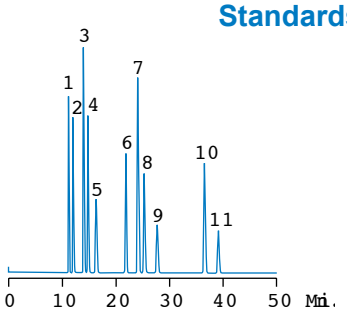
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	30
%B:	20	29

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Vegetable Fermentation Broth Standards

CHROM 10004



1. iso-erythritol
2. Fructose
3. Sorbitol
4. Mannitol
5. Glucose
6. Inositol
7. Sucrose
8. Maltitol
9. Maltose
10. Raffinose
11. Maltotriose

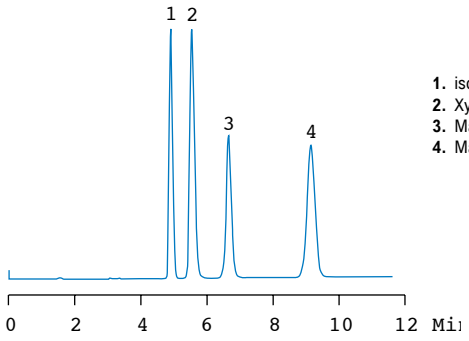
Column: Prevail™ Carbohydrate ES, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** 0.04% NH_4OH
Gradient:

Time:	0	60
%B:	17	35

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Sugar Alcohols

CHROM 9352

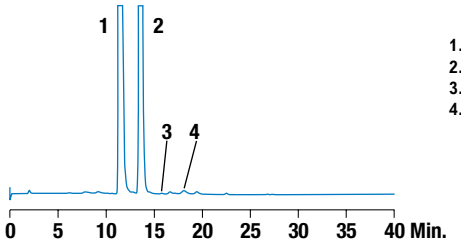


1. iso-Erythritol
2. Xylitol
3. Mannitol
4. Maltitol

Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: Acetonitrile:Water (70:30)
Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Cough Syrup

CHROM 10556



1. Fructose
2. Glucose
3. Sucrose
4. Maltose

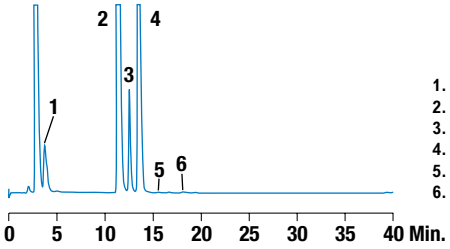
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	5	10	15	25
%B:	15	15	25	25	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Robitussin® Flu Syrup

CHROM 10557



1. Sucralose
2. Fructose
3. Sorbitol
4. Glucose
5. Sucrose
6. Maltose

Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

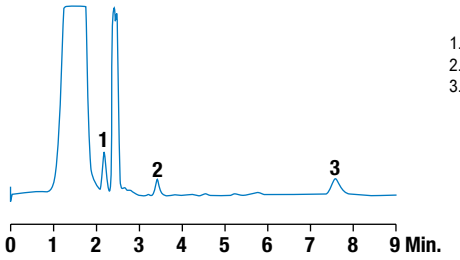
Time:	0	5	10	15	25
%B:	15	15	25	25	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Spiked Urine

Chromatogram courtesy of Rosemary Rose, Monash University, Dept. of Gastroenterology

CHROM 10584

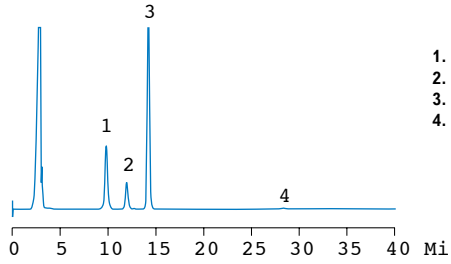


1. Sucralose
2. Rhamnose
3. Cellobiose

Column: Prevail™ Carbohydrate ES, 5µm, 150 x 4.6mm, (Part No. **35102**)
Mobile Phase: Acetonitrile:Water (70:30)
Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Tobacco

CHROM 10526



1. Fructose
2. Glucose
3. Sucrose
4. Unknown

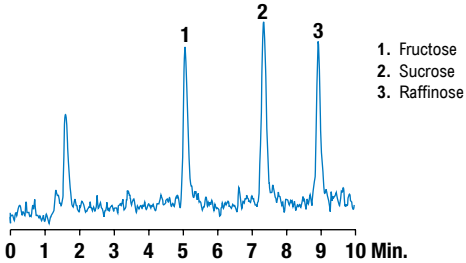
Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	5	10	15	25
%B:	20	20	30	30	50

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Low Level Detection - 10ng on Column

CHROM 10498



1. Fructose
2. Sucrose
3. Raffinose

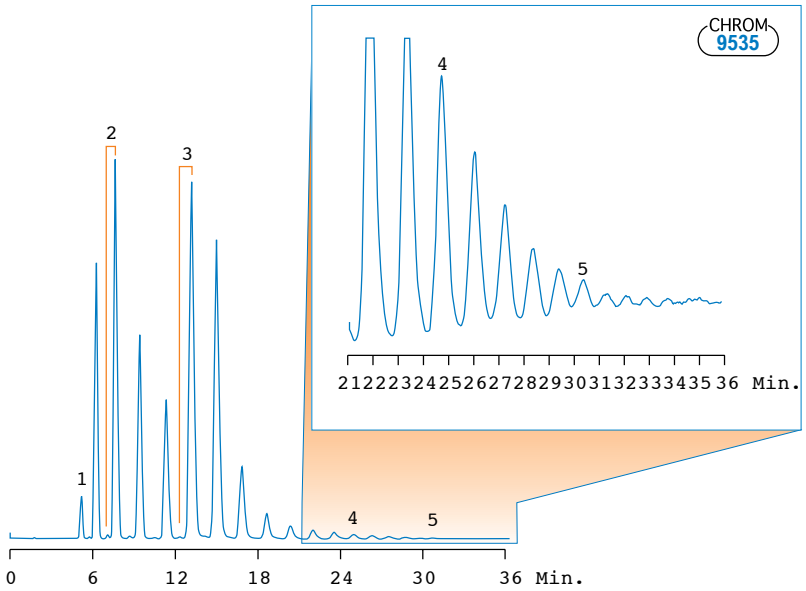
Column: Prevail™ Carbohydrate ES, 5µm, 150 x 4.6mm, (Part No. **35102**)
Mobile Phase: **A:** Water **B:** Acetonitrile
Gradient:

Time:	0	10	11	14
%B:	80	50	80	80

Flow Rate: 1.0 mL/min
Detector: ELSD
Column Temp: Ambient

Glucooligosaccharides in Maltodextrin

CHROM 9534



1. Glucose (DP1)
2. DP3 (1,6-alpha; 1,4-alpha)
3. DP6 (1,6-alpha; 1,4-alpha)
4. DP13
5. DP18

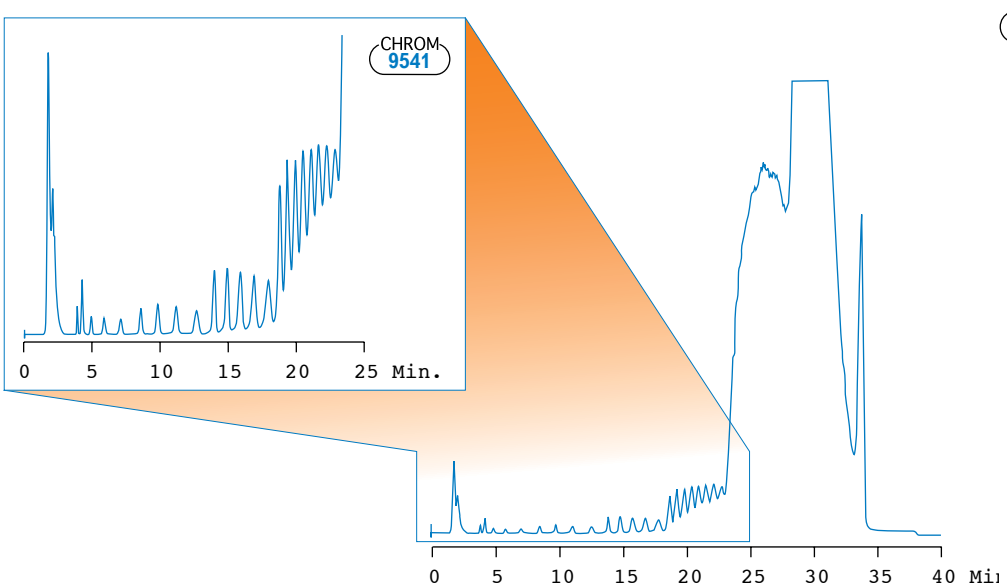
Column: Prevail™ Carbohydrate ES, 5µm 250 x 4.6mm (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient:

Time:	0	50	60
%B:	35	60	60

Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Partially Hydrolyzed Dextran

CHROM 9540



Column: Prevail™ Carbohydrate ES, 5µm, 250 x 4.6mm, (Part No. **35101**)
Mobile Phase: **A:** Acetonitrile **B:** Water
Gradient: Step (5 min. hold at 40% B, 5% B steps every 5 min. to 70% B)
Flow Rate: 1.0mL/min
Detector: ELSD
Column Temp: Ambient

Prevail™ Carbohydrate ES HPLC Columns

Prevail™ Carbohydrate ES HPLC Columns are available in a variety of formats. Choose from analytical, solvent reducing, and preparative formats.

PREVAIL™ HPLC COLUMNS

Packing	Format	I.D. x Length	Part No.
Carbohydrate ES, 5µm	LC/MS	2.0 x 150mm	**35116**
	Analytical	4.6 x 150mm	**35102**
	Analytical	4.6 x 250mm	**35101**
	Rocket™	7.0 x 53mm	**35104**
	Prep	7.0 x 100mm	**35103**
	Prep	10 x 250mm	**35115**

High-Speed Rocket™ Columns Reduce Analysis Time and Solvent Consumption

Separations normally performed on conventional 150mm x 4.6mm columns run faster on Rocket™ Columns and use less mobile phase, saving time and money. The column design allows higher flow rates with reduced back pressures while maintaining good plate counts. At a flow rate of 3mL/min, a simple sugar separation can be accomplished in less than 5 minutes on a 53 x 7mm Rocket™ Column.

Actual Size (33 x 7mm)

PREVAIL™ ALL-GUARD™ CARTRIDGES*

Packing	I.D. x Length	Qty	Part No.
Carbohydrate ES	4.6 x 7.5mm	3	**96435**
All-Guard™ Guard Cartridge Holder		1	**80101**

*All-Guard™ Holder required

PREVAIL™ PREP-GUARDS*

Packing	I.D. x Length	Qty	Part No.
Carbohydrate ES	7.6 x 50mm	ea	35105
Direct-Connect™ Column Coupler		ea	28195

*Direct-Connect™ Column Coupler required

To learn more about Alltech® ELSDs, call 1-800-255-8324 or visit our website: www.discoverysciences.com/ELSD

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10/08 #467B