Foresight™ Chromatography Media Columns 1 and 5 ml

Instruction Manual

Catalog Numbers

732-4720	732-4730
732-4740	732-4750
732-4721	732-4731
732-4741	732-4751
732-4722	732-4732
732-4742	732-4752
732-4735	732-4736
732-4755	732-4756

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Section 1
Introduction

Foresight™ chromatography media columns have a robust design that provides convenience and reliability for process scientists. Foresight columns may be used for qualitative media screening experiments and/or small scale methods development. Through design of experiments (DoE) utilizing Foresight columns, an operational window can be defined to aid in developing a quality by design (QbD) process. The design of the column ensures leak-free operation at pressures up to 30 bar. The columns allow faster, more convenient experimental setup.

The design of the Foresight column offers:

1. **Convenience**
   The prepacked column is ready for equilibration in the buffer of choice. Bed heights are available in ratios that allow straightforward comparative evaluation.
2. **Reliability**
   Each column is individually packed under optimum compression, ensuring consistent experimental results.

Foresight columns are available in a variety of chromatography modes including anion, cation, and mixed-mode. See Section 5, Ordering Information, for the complete Foresight column product line.

Table 1. Foresight chromatography media column properties.

Connectors	10–32 fittings	
Column material	Polyethylene (HDPE)	
Frit material	Polypropylene/polyethylene	
Shipping conditions		
Ion exchangers	20% ethanol + 0.15 M NaCl	
CHT™	Dry	
Nuvia™ cPrime™	20% ethanol	
Storage	20% ethanol or 0.1 N NaOH	
Autoclavability	Not autoclavable	
	1 ml	**5 ml**
Dimensions		
Inner diameter (ID)	8 mm	8 mm
Length	20 mm	100 mm
Max pressure	20 bar (290 psi)	30 bar (435 psi)
Recommended flow rates	0.5–1 ml/min 59.5–119 cm/hr	2.5–5 ml/min 297.5–595 cm/hr
Max flow rate	8 ml/min 952 cm/hr	8 ml/min 952 cm/hr

Table 2. Chromatography media specifications.

Specifications for all chromatography media may be found by visiting www.bio-rad.com/process and downloading their corresponding instruction manuals.

Chromatography Media	Mode	Instruction Manual Literature #
UNOsphere™ Q	Strong anion	4110109
UNOsphere S	Strong cation	4110109
UNOsphere Rapid S	Strong cation	10010339
Nuvia Q	Strong anion	10018215
Nuvia S	Strong cation	10018215
Nuvia cPrime	Mixed-mode hydrophobic-cationic	10023853
CHT type I-40	Mixed mode metal affinity-cationic	6086
CHT type II-40	Mixed-mode metal affinity-cationic	6086

Section 2
Connecting to Bio-Rad's Low-Pressure Chromatography Systems and Other Liquid Chromatography Systems

Foresight™ columns have standard HPLC connections (M10–32 UNF for 1/16" tubing) which make them compatible with many liquid chromatography systems.

2.1 Preparation of Column Tubings

2.1.1 For Bio-Rad Laboratories' BioLogic Duoflow™ system, 1/16" OD inlet and outlet tubings are recommended. If the DuoFlow system tubings have male 1/4–28 fittings attached, use male 10–32 to female 1/4–28 adaptors (catalog #750-0564) to connect column to DuoFlow system. Alternatively, use long 10–32 fittings (catalog #760-1311) to attach to the tubing.

2.1.2 For GE Healthcare ÄKTA systems, 1/16" OD inlet and outlet tubings are recommended. If the system tubings have male 1/16" fittings, these will attach directly to the column. If tubing fittings are required, use long 10–32 fittings (catalog #760-1311) to attach to the tubings.

2.1.3 For BioLogic™ LP system, 0.8 or 1.6 mm tubing is recommended. Use fitting from IDEX Health & Science P-642 female luer to male 10–32 adaptor to connect to column. Use either 0.8 mm barb to male luer (catalog #731-8224), 1.6 mm barb to male luer (catalog #731-8225), or 3.2 barb to male luer (catalog #731-8226) to attach to the respectively-sized tubing on the BioLogic LP system.

2.2 Connecting the Column to a System

2.2.1 Mount the column in a vertical position with the arrow pointed downward.

2.2.2 Remove the stop plugs from the top and bottom of the column.

2.2.3 Connect the inlet tubing to the solvent delivery system and with a flow rate of 0.2 ml/min, connect the inlet tubing to the column. Push the tubing in until it bottoms firmly and tighten the 1/16" fingertight male fitting to the female end piece on the column.

2.2.4 When the solvent is flowing freely from the outlet end of the column, connect the outlet tubing to the column by pushing the tubing in until it bottoms firmly and tighten the 1/16" fingertight male fitting to the female end piece on the column. Ensure that the solvent is present in the column assembly, and then connect it to the detector of the chromatography system.

Section 3
Preparing a Column for Use

Foresight™ columns may be shipped to you containing 20% ethanol, 20% ethanol + 0.15 M NaCl (v/v), or dry, depending on the chromatography media. All columns are ready to use after equilibrating the columns in the buffer of choice. To perform a buffer exchange, connect the column to a liquid chromatography system or peristaltic pump and condition it as follows:

1. Set the pump flow rate to 0.5–1 ml/min (59.5–119 cm/hr) for the 1 ml column or 2.5–5 ml/min (297.5–595 cm/hr) for the 5 ml column.
2. Wash the column with degassed low-salt buffer for 2 column volumes. Wash with degassed high-salt buffer for 5 column volumes.
3. Equilibrate with low-salt buffer for 5 column volumes.
4. Reduce the flow rate to the rate that will be used in the purification protocol.

3.1 Sample Preparation

Correct pH and ionic strength are necessary for consistent and reproducible results. Sample can be exchanged into the starting buffer or diluted to the starting buffer concentration. This can be achieved by diluting the sample to the ionic strength of the starting buffer, dialyzing against the starting buffer, or exchanging into the starting buffer. Buffer exchange can be accomplished using a number of products (Table 3). The choice of product will depend on sample volume. Filter all samples through a 0.45 µm filter prior to column application.

Table 3. Products for buffer exchange.

Sample Volume	Product	MW Cutoff	Catalog
50–100 µl	Bio-Spin® columns	6 kD	732-6002
	Bio-Spin 30 columns	30 kD	732-6006
0.1–3 ml	Bio-Scale™ mini cartriges	6 kD	732-5304
Up to 3 ml	Econo-Pac® 10DG6 columns	6 kD	732-2010
Unlimited	Bio-Gel® P-6DG gel	6 kD	150-0738

3.2 General Purification Protocol

Recommendations on general purification protocols may be found for each chromatography media in its corresponding instruction manual. Please refer to Table 2 for literature number, and download at www.bio-rad.com/process. For best results and increased column life, samples and buffers should be degassed and filtered through a 0.45 µm filter.

3.3 Purification Scale-Up

For quick scale-up, two or three columns of the same type can be connected in series. Backpressure will increase with columns in series, so care should be taken to maintain pressures ≤ 20 bar for 1ml and ≤ 30 bar for 5 ml. Foresight columns are available in 1 and 5 ml column formats. All chromatography media are designed for process scale and are available in larger bulk quantities for uses from preclinical to commercial. All media are fully supported with regulatory support files. In addition, Bio-Rad carries an extensive line of empty chromatography columns for use in laboratory and process scale applications.

Section 4
Cleaning-in-Place (CIP) and Sanitation

If a column no longer yields reproducible results, the media may require thorough CIP and sanitation to remove strongly bound contaminants. Acceptable CIP agents include 25% acetic acid, 8 M urea, 1% Triton X-100, 6 M potassium thiocyanate, 70% ethanol, 30% isopropyl alcohol, 1 N NaOH, and 6 M guanidine hydrochloride.

1. Sanitize the support in the column with 2–4 bed volumes of 1.0 N NaOH while maintaining a contact time of at least 40 min.
2. To reequilibrate the column, wash it with 2–4 bed volumes of 0.5–2 M NaCl solution (containing 50–100 mM buffer).
3. If lipid removal is required, wash the column with a 20–70% ethanol solution.

4.1 Autoclaving

Foresight™ columns are not autoclavable.

4.2 Storage

After washing the columns with deionized water, Foresight columns should be purged, stored in the recommended solution for the specific media, and capped for extended storage.

Section 5
Ordering Information

Foresight™ Chromatography Media Columns

Product	Mode	Catalog #
Foresight™ Nuvia™ S Column, 1 ml	Strong Cation	732-4720
Foresight Nuvia S Column, 5 ml	Strong Cation	732-4740
Foresight Nuvia Q Column, 1 ml	Strong Anion	732-4721
Foresight Nuvia Q Column, 5 ml	Strong Anion	732-4741
Foresight™ Nuvia™ cPrime™ Column, 1 ml	Mixed Mode (Hydrophobic-Cationic)	732-4722
Foresight Nuvia cPrime Column, 5 ml	Mixed Mode (Hydrophobic-Cationic)	732-4742
Foresight™ UNOsphere™ S Column, 1 ml	Strong Cation	732-4730
Foresight UNOsphere S Column, 5 ml	Strong Cation	732-4750
Foresight UNOsphere rS Column, 1 ml	Strong Cation	732-4731
Foresight UNOsphere rS Column, 5 ml	Strong Cation	732-4751
Foresight UNOsphere Q Column, 1 ml	Strong Anion	732-4732
Foresight UNOsphere Q Column, 5 ml	Strong Anion	732-4752
Foresight™ CHT™ Type I, 40 µm Column, 1 ml	Mixed Mode (Metal Affinity-Cationic)	732-4735
Foresight CHT Type I, 40 µm Column, 5 ml	Mixed Mode (Metal Affinity-Cationic)	732-4755
Foresight CHT Type II, 40 µm Column, 1 ml	Mixed Mode (Metal Affinity-Cationic)	732-4736
Foresight CHT Type II, 40 µm Column, 5 ml	Mixed Mode (Metal Affinity-Cationic)	732-4756

Foresight Chromatography Media Filter Plates

Product	Mode	Catalog #
Foresight™ Nuvia™ S Filter Plate, 20 µl	Strong Cation	732-4701
Foresight Nuvia Q Filter Plate, 20 µl	Strong Anion	732-4703
Foresight™ Nuvia™ cPrime™ Filter Plate, 20 µl	Mixed Mode (Hydrophobic-Cationic)	732-4705
Foresight™ UNOsphere™ S Filter Plate, 20 µl	Strong Cation	732-4710
Foresight UNOsphere rS Filter Plate, 20 µl	Strong Cation	732-4712
Foresight UNOsphere Q Filter Plate, 20 µl	Strong Anion	732-4714
Foresight™ CHT Type™ I, 40 µm Filter Plate, 20 µl	Mixed Mode (Metal Affinity-Cationic)	732-4716
Foresight CHT Type II, 40 µm Filter Plate, 20 µl	Mixed Mode (Metal Affinity-Cationic)	732-4718

Foresight™ RoboColumns® Units

Product	Mode	Catalog #
Foresight™ Nuvia™ S RoboColumn, 200 µl	Strong Cation	732-4801
Foresight Nuvia S RoboColumn, 600 µl	Strong Cation	732-4802
Foresight Nuvia Q RoboColumn, 200 µl	Strong Anion	732-4804
Foresight Nuvia Q RoboColumn, 600 µl	Strong Anion	732-4805
Foresight™ Nuvia™ cPrime™ RoboColumn, 200 µl	Mixed Mode (Hydrophobic-Cationic)	732-4807
Foresight Nuvia cPrime RoboColumn, 600 µl	Mixed Mode (Hydrophobic-Cationic)	732-4808
Foresight™ UNOsphere™ S RoboColumn, 200 µl	Strong Cation	732-4813
Foresight UNOsphere S RoboColumn, 600 µl	Strong Cation	732-4814
Foresight UNOsphere rS RoboColumn, 200 µl	Strong Cation	732-4816
Foresight UNOsphere rS RoboColumn, 600 µl	Strong Cation	732-4817
Foresight UNOsphere Q RoboColumn, 200 µl	Strong Anion	732-4819
Foresight UNOsphere Q RoboColumn, 600 µl	Strong Anion	732-4820
Foresight™ CHT™ Type I, 40 µm RoboColumn, 200 µl	Mixed Mode (Metal Affinity-Cationic)	732-4822
Foresight CHT Type I, 40 µm RoboColumn, 600 µl	Mixed Mode (Metal Affinity-Cationic)	732-4823
Foresight CHT Type II 40 µm RoboColumn, 200 µl	Mixed Mode (Metal Affinity-Cationic)	732-4825
Foresight CHT Type II 40 µm RoboColumn, 600 µl	Mixed Mode (Metal Affinity-Cationic)	732-4826

Fittings Kits

Catalog #	Description
731-8226	3.2 mm Barb to Male Luer, pkg of 25
731-8225	1.6 mm Barb to Male Luer, pkg of 25
731-8224	0.8 mm Barb to Male Luer, pkg of 25
750-0564	HPLC Column to Biologic System Adaptors
760-1311	Long Fingertight Fittings, 10-32 x 1.03”

Section 6
Related Reading

Harris ELV and Angal S (1989). Protein Purification Methods: A Practical Approach (Oxford: IRL Press).

Scopes RK (1987). Protein Purification: Principles and Practice, second edition (New York: Springer-Verlag).

Snyder LR and Kirkland JJ (1979). Introduction to Modern Liquid Chromatography, second edition (New York: Wiley).

Gagnon P (1997). Avoiding instrument-associated aberrations in purification scale-up and scale-down, BioPharm 10, 42–45.

Fittings to Bio-Rad Laboratories LP system: http://www.idex-hs.com/Default.aspx

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