

Agilent Prep LC Columns - High Performance Preparative Columns for Maximum Sample Purification

Technical Overview

Highlights

- High loadability for maximum sample purification
- Easy scalability from 4.6-mm ID to 50-mm ID for rapid method development
- High-Throughput 21.2-mm ID cartridges for fast purification
- Exceptional column stability from pH 2-10
- Ideal for use with the Agilent Purification System

Agilent Prep LC Columns - A New Choice for Maximum Sample Purification

High quality HPLC sample purification is critical in expanding drug discovery and drug development programs. Agilent Prep LC columns, available in both C18 and silica, provide high loadability for maximum sample purification with easy scalability to assure isolation and purification of milligrams to grams of desired drug compounds. At the same time, Agilent Prep LC columns provide the stability, high performance and flexibility to meet the most demanding purification needs.

Superior Loadability over a Wide pH Range

Sample loadability is determined by many factors, including sample type (acidic, basic or neutral), resolution and mobile phase pH. Therefore, the best preparative columns provide excellent performance under a wide range of conditions. The new Agilent Prep columns provide superior loadability under low (pH 2) and high (pH 10) pH conditions with basic compounds - the most difficult sample type. See Figure 1.

Agilent Prep columns allow for a greater load with difficult basic compounds. At pH 10, the best competitive columns allowed for only 78% of the sample load of the Agilent Prep-C18 column before the peak width broadened substantially. At pH 2, the Agilent Prep-C18 column provided more than double the sample load for the same basic compounds. While these comparisons were made using analytical (4.6 mm ID) columns, the results scaleup to larger ID columns. Agilent Prep columns provide superior loadability in any dimension.



Figure 1. Loadability comparison of the basic compound, oxybutynin, at pH 2 and 10 in DMSO, using Agilent Prep-C18 and two competitive columns.



This superior loadability is achieved with highpurity, high surface-area silica using a C18 bonded phase suitable for purification of a wide range of nonpolar and polar compounds.

Table 1 compares the loadability of methyl- and ethyl benzoate on an Agilent Prep-C18 column against two competitors.

Table 1. Loadability Comparison

Column	mg loaded on at Rs = 1.25	mg loaded per g packing	mg loadable in 50 × 250-mm column		
Agilent Prep-C18	4.80	3.47	1735		
Competitor K C18	4.45	2.68	1340		
Competitor W C18	3.88	2.32	1160		
Mobile phases:	$A=H_2O$				
	B=Methanol, isocratic, 70.5%–78% B (initial resolution = 1.8)				
Column temperature:	30 °C				
Injection:	125 μL of each dilution				
Sample:	100 mg/mL each of methyl benzoate and ethyl benzoate in 90% methanol				

This unique bonded phase also provides outstanding peak shape with basic compounds allowing very high purity sample isolation. See Figure 2.



Agilent Prep-C18

Competitor W-C18

Figure 2. Loadability comparisons for the basic compounds doxepin and amitriptyline, using Agilent Prep-C18 and a competitive column.

Easy Scalability for Rapid Method Development

Column dimensions are available for preparative samples ranging from high-throughput drug discovery samples to general lab scale prep samples. Preparative columns are available in 21.2, 30, and 50-mm ID with lengths ranging from 50–250 mm, many with both 5- and 10-µm particle sizes standard. This wide variety of dimensions allows for optimizing the separation and maximizing the sample load on a scalar (analytical 4.6-mm ID) column before selecting the size of the preparative column needed. Your separation and results will have the same high quality as you choose larger column dimensions for greater sample purification. See the example shown in Figure 3.



Figure 3. An example of the excellent scalability of the Agilent Prep-C18 column, under nonoverload conditions, making method transfer simple and predictable.

For more rapid method development, one can also immediately select a high performance preparative column based on estimated sample loads, as shown in Table 2.

Table 2.	Typical Flow Rates and Column Loads Based on
	Column Size and Separation

		Separation type (small molecules)		
Column ID (mm)	Normalized flow rate (mL/min)	Easy (α >1.5) (mg)	Difficult (a >1.2–1.5) (mg)	
4.6	1	3–15	0.5–3	
21.2	20	70–400	20–70	
30	40	140-800	40–140	
50	100	400–2000	100–400	

Rapid, High-Throughput Purification

High-Throughput drug purification requires new column choices for preparative separations. Shorter columns (50–100 mm long) and smaller (<10 micron) particles are ideal. Agilent Prep 21.2-mm ID columns are available with Agilent's Preparative Cartridge Hardware (PrepHT). This reliable cartridge hardware makes it easy to use short (50- and 100-mm long) columns for rapid sample purification and then quickly change to a longer column length to conveniently increase sample load. For even larger increases in sample load, 30-mm and 50-mm ID columns with fixed end fittings can be used. Cartridge columns (21.2-mm ID) are also available in longer 150- and 250-mm lengths for more complex samples, such as mixtures of natural compounds. Longer (150, 250 mm) 30- and 50-mm ID columns are available for product isolation. Guard columns are easily integrated onto any of these columns, providing superior protection of the prep column with complex samples.

Exceptional Column Stability

Increased throughput requires improved preparative column stability for reliable and reproducible results. Agilent Prep columns demonstrate excellent stability for more than 1000 injections under a variety of solvent conditions. This is demonstrated using the Agilent Prep-C18, 21.2 mm \times 150 mm, 10-µm column in Figure 4. This allows for increased productivity by providing the stability to run unattended for long periods of time.



No changes in efficiency and tailing over 1000+ injections indicates the column is stable and the packed bed is not compressing.

Figure 4. Agilent Prep-C18 column mechanical stability over 1000+ injections, using many solvent types.

Flexibility for All Your Purification Needs

Whether it is improving loadability by using high pH or improving throughput and efficiency with a $5-\mu m$ particle size and short column length, Agilent Prep columns provide the flexibility for your demanding purification samples. These reliable

preparative columns are available in a wide range of dimensions to meet all of your preparative needs. See Tables 3 and 4. Agilent Prep columns are also the best choice when using the Agilent Purification System. Now Agilent provides complete solutions for the isolation and purification of the widest range of compounds.

Table 3. Agilent Prep Columns - Specifications

Bonded phase	Pore size	Surface area	Temp. limits	pH Range	Endcapped	Carbon load
C18	100 Å	400 m²/g	60 °C*	2–10	Yes	24%
Silica	100 Å	400 m²/g	**	1–8	N/A	N/A

N/A Not Applicable

*Temperature limits are 60 °C up to pH 8, 40 °C for pH 8–10.

**Temperature limits for bare silica are determined by the temperature of the mobile phase.

Column description	Size (mm)	Particle size (µm)	C18	Silica
Scalar	4.6×250	5	440905-902	440905-901
Scalar	4.6×150	5	443905-902	443905-901
Scalar	4.6×100	5	449905-902	449905-901
Scalar	4.6 imes 50	5	446905-902	446905-901
PrepHT cartridge columns (require	hardware kit 820400	-901)		
PrepHT	21.2 × 250	10	410910-102	410910-101
PrepHT	21.2 × 150	10	413910-102	413910-101
PrepHT	21.2 × 150	5	443905-102	443905-101
PrepHT	21.2 × 100	5	449905-102	449905-101
PrepHT	21.2×50	5	446905-102	446905-101
Standard fittings (no hardware requ	iired)			
Prep 30	30 imes 250	10	410910-302	410910-301
Prep 30	30 imes 150	10	413910-302	413910-301
Prep 30	30 imes 100	10	419910-302	419910-301
Prep 30	30 imes 100	5	449905-302	449905-301
Prep 30	30 imes 50	5	446905-302	446905-301
Prep 50	50 imes 250	10	410910-502	410910-501
Prep 50	50 imes 150	10	413910-502	413910-501
Prep 50	50 imes 100	10	419910-502	419910-501
Prep 50	50 imes 100	5	449905-502	449905-501
Prep 50	50 imes 50	5	446905-502	446905-501
PrepHT Column Hardware Kit	_	_	820400-901	820400-901
PrepHT Guard Cartridges, 2/pk	21.2×10	10	420212-902	420212-901
Prep HT Guard Hardware	_	_	820444-901	820444-901
Prep External Guard Hardware	_	-	420420-901	420420-901
Bulk packing	1 kg	10	420910-902	420910-901

Table 4. Agilent Prep Columns - Ordering Information

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All PrepHT cartridge columns require hardware kit 820400-901. If a guard column is desired for the 21.2-mm ID columns, the PrepHT Guard Hardware Kit, 820444-901, is also required.



PrepHT Guard Hardware Kit, 820444-901

If the guard column is used on a 30-mm or 50-mm ID column the External Guard column hardware kit, 420420-901, is required



Prep External Guard Hardware Kit, 420420-901

Agilent Prep-C18 30 \times 250 mm, 10 $\mu\text{m},$ p/n 410910-302

For More Information

For more information on our products and services, visit our Web site at www.agilent.com/chem.

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Printed in the USA July 9, 2004 5989-1269EN

