

HPLC Column Selection: Small Molecules (<2000 mw)

This chart is meant to be a starting point for choosing an HPLC column. In some cases, more than one column can be used for an analysis. Although general rules apply, you should first consult the technical literature, official methodology, or our Technical Service chemists for definitive recommendations.

For column choices for peptides, proteins, and other biopolymers, see pages 54-55.

ANALYTE CHARACTERISTICS		TECHNIQUE	ANALYTE CLASS	COLUMN	
water soluble	nonionic	reversed phase	GENERAL (pH 2-7.5)	Discovery C18, C8, RP-AmideC16; Discovery Cyano, Discovery HS C18, HS F5, HS PEG	
			GENERAL (pH 2-13)	SUPELCOGEL ODP-50, TPR-100	
			acids, organic	Discovery C18, C8, RP-AmideC16	
			acids, amino	Discovery C18, RP-AmideC16; SUPELCOGEL LC-DABS	
			amines, organic	Discovery C18, RP-AmideC16; Discovery Cyano, Discovery HS F5; SUPELCOGEL TPR-100	
			amines, quaternary (pH≥7.5)	SUPELCOGEL TPR-100, ODP-50	
			chelating compounds	Discovery C18, C8, RP-AmideC16; SUPELCOGEL TPR-100, ODP-50	
			drugs, basic	Discovery C18, C8, RP-AmideC16; Discovery Cyano, Discovery HS F5	
			drugs in serum	Hisep; Discovery C18, RP-AmideC16, C8; Discovery HS F5	
			explosives	Discovery C18, C8, RP-AmideC16; Discovery Cyano	
			nucleosides	SUPELCOGEL LC-18-S; Discovery C18, RP-AmideC16	
			nucleotides	SUPELCOGEL LC-18-T; Discovery C18, RP-AmideC16	
	peptides	Discovery C18, C8, RP-AmideC16			
	sugars	SUPELCOGEL LC-NH ₂			
	taxol, taxanes	SUPELCOGEL LC-F			
	tricyclic antidepressants	SUPELCOGEL LC-PCN; Discovery C8; Discovery Cyano			
	vitamins, water soluble	Discovery C18, C8, RP-AmideC16			
	water soluble	hydrophobic interaction	size exclusion	peptides	TSK-GEL
oligosaccharides				SUPELCOGEL Ag1, Ag2; TSK-GEL Oligo-PW	
polymers/oligomers, hydrophilic				TSK-GEL PW _{XL}	
ionic	ion exchange	ion pairing	anions, organic (- charge)	SUPELCOGEL SAX1	
			cations, organic (+ charge)	SUPELCOGEL LC-SCX	
	ion exclusion	ion exclusion	acids, organic	Discovery C18, Discovery HS C18	
			amines, organic	Discovery C18, Discovery HS C18	
organic soluble	normal phase	normal phase	GENERAL	Discovery Cyano, LC-NH ₂ -NP, LC-Si, Discovery HS PEG	
			afatoxins	SUPELCOGEL LC-Si	
			steroids	SUPELCOGEL LC-Diol	
			vitamins, fat soluble	SUPELCOGEL LC-NH ₂ -NP	
	organic soluble	reversed phase	reversed phase	fatty acids	SUPELCOGEL LC-18
				polyaromatic hydrocarbons	SUPELCOGEL LC-PAH
				polymers, hydrophobic	SUPELCOGEL LC-18
				triglycerides	SUPELCOGEL LC-18
				vitamins, fat soluble	Discovery C18, C8, RP-AmideC16, SUPELCOGEL LC-8
	organic soluble	size exclusion	size exclusion	polymers/oligomers, hydrophilic	TSK-GEL H _{HR}

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

Take Advantage of the Discovery Suite of Reversed-Phases

Whether you're developing a separation that must be perfect, or you're just not satisfied with your reversed-phase separation...

Turn to Discovery!

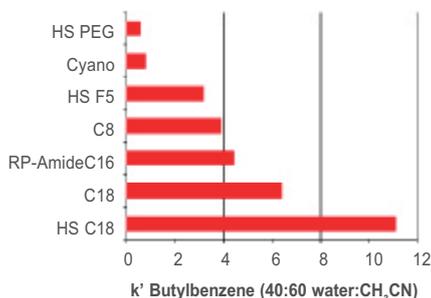
a suite of HPLC columns featuring functionalized reversed-phases designed to provide differentiated separations vs. C18 based on unique combinations of polar and hydrophobic retention.

The Discovery suite of reversed-phases enables you to optimize your separation..., e.g.

- retention
- selectivity
- resolution
- analysis time

...while minimizing method development time.

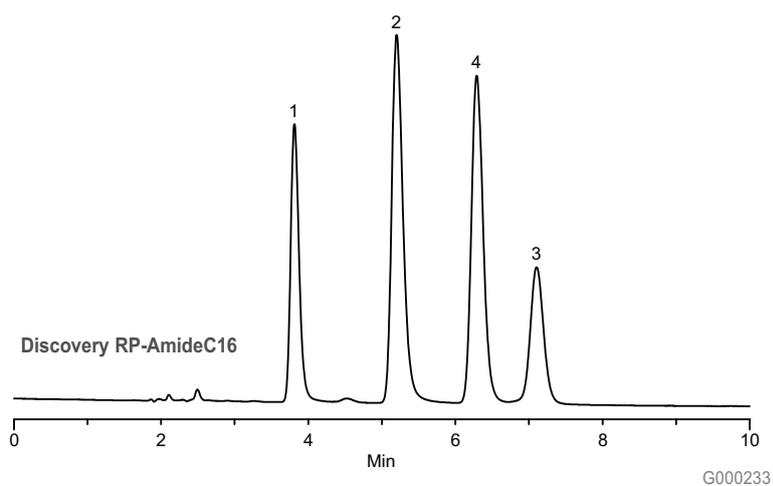
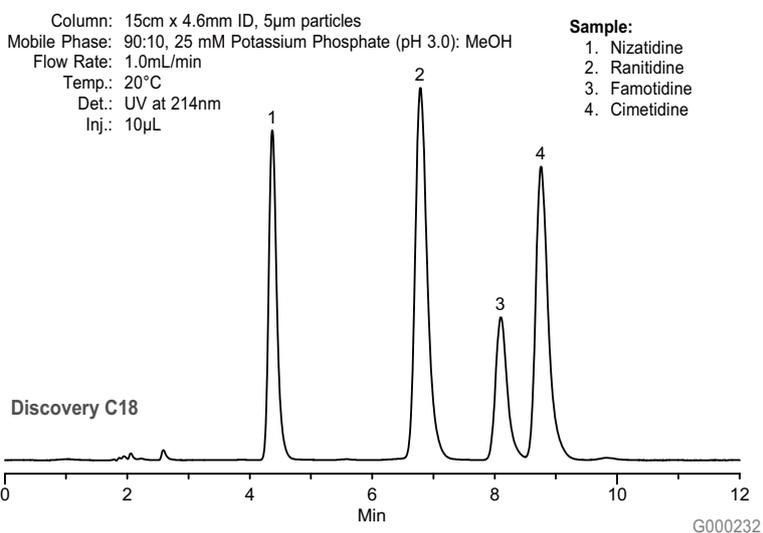
Hydrophobicity: Discovery Reversed-Phases



Discovery RP-AmideC16 Delivers Better Peak Spacing and Faster Analysis!

Scientists have long known polar embedded phases such as Discovery RP-AmideC16 provide unique retention and selectivity compared to C18's. Taking advantage of Discovery RP-AmideC16's unique retention and selectivity results in a better separation vs. C18.

- Unique selectivity - peak reversal of compounds 3 and 4
- Better peak spacing - less dead space between peaks 1 and 2
- Higher assay throughput - 20% reduction in analysis time
- Improved resolution of critical peak pair 3 and 4



HPLC: Small Molecules

Rediscover Method Development

US \$

Deliver Better Separations in Less Time

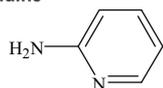
Unique retention and selectivity of Discovery HS F5 enables rapid development of simple impurity assay where C18 fails!

Impurity methods requiring retention and resolution of vastly differing analytes may not be suitably obtained using simple C18-based systems. By simply changing the stationary phase the method development scientist can avoid:

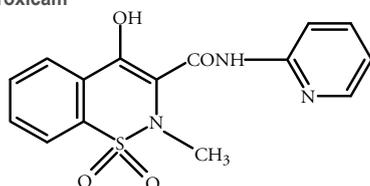
- complicated or forbidden gradients
- complex mobile phases
- long, drawn-out method development

On Discovery HS F5, it took just a few hours to develop an excellent separation.

2-Aminopyridine



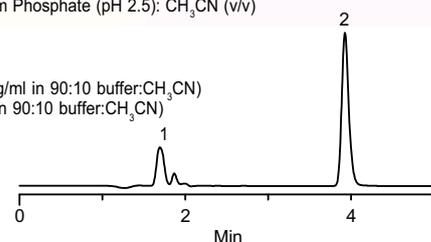
Piroxicam



2-Aminopyridine (2-AMP) is Unretained on C18 Under Mobile Phase Conditions Used to Assay Piroxicam

Column: Discovery C18 15cm x 4.6mm ID, 5µm particles
 Mobile Phase: 45:55, 10 mM Potassium Phosphate (pH 2.5): CH₃CN (v/v)
 Flow Rate: 1.0mL/min
 Det.: UV at 220nm
 Inj.: 5µL
 Sample: 1. 2-Aminopyridine (10µg/ml in 90:10 buffer:CH₃CN)
 2. Piroxicam (100µg/ml in 90:10 buffer:CH₃CN)

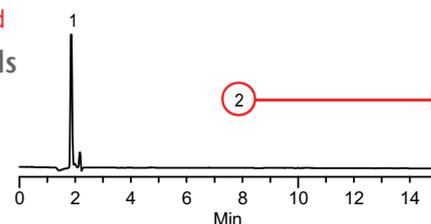
1. 2-Aminopyridine
2. Piroxicam



Decreasing the % Acetonitrile Results in Excessive Piroxicam Retention and 2-AMP is Still Unretained

C18 Fails

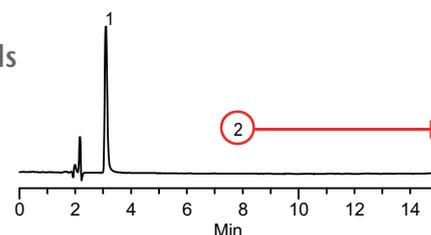
Same buffer but with lower organic:
 85:15, 10 mM Potassium Phosphate (pH 2.5): CH₃CN (v/v)



Increasing pH to 6.8 Retains the 2-AMP but Piroxicam Retention is Still Excessive

C18 Fails

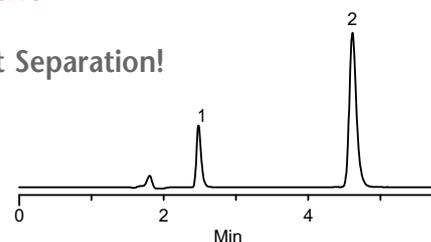
Same %organic, but changing the pH to 6.8: 85:15, 10 mM Potassium Phosphate (pH 6.8): CH₃CN (v/v)



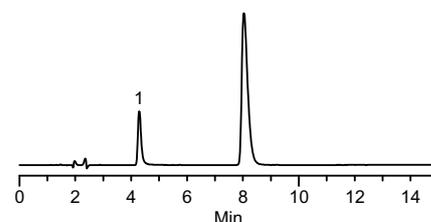
The Unique Retention and Selectivity of Discovery HS F5 Produces Excellent Separation at Both pH's

F5 Delivers Excellent Separation!

85:15, 10 mM Potassium Phosphate (pH 2.5): CH₃CN (v/v)



85:15, 10 mM Potassium Phosphate (pH 6.8): CH₃CN (v/v)



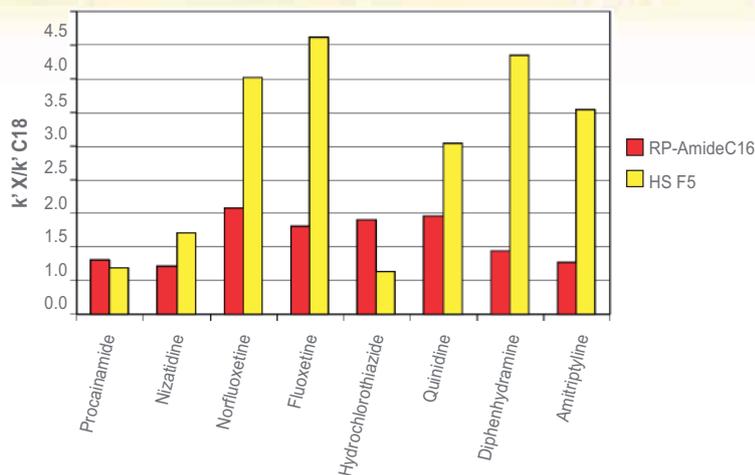
Different Retention is a General and Valuable Characteristic of Functionalized Reversed-Phases

Unique retention vs. C18

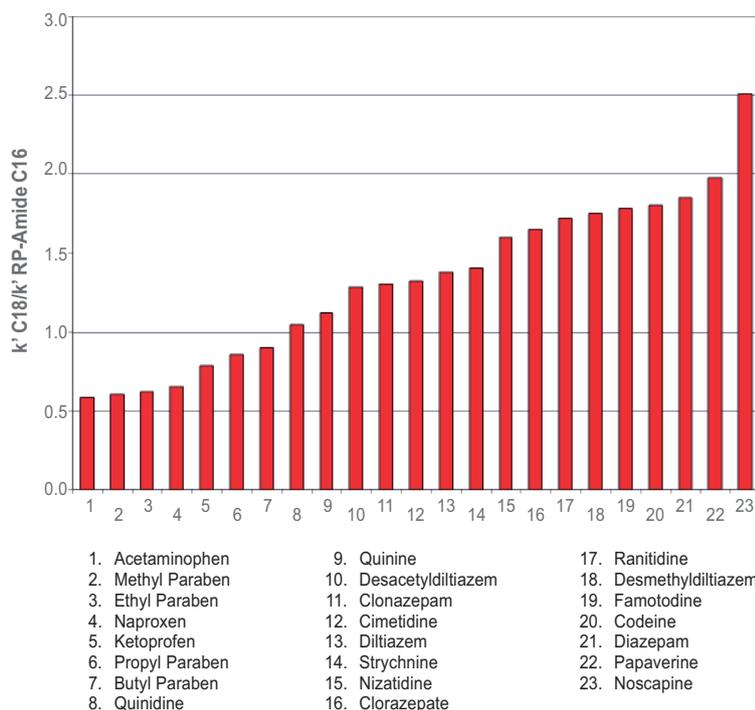
Retention of several basic drugs is compared on C18, RP-AmideC16 and F5. (F5 mobile phase %organic is +30% vs. C18 and RP-AmideC16.)

- Both F5 and RP-AmideC16 deliver valuable, different retention compared to C18.
- F5 is significantly more retentive for most compounds tested vs. both C18 and RP-AmideC16
- Retention on functionalized reversed-phases is almost always different than retention on C18!

Retention on RP-AmideC16 and F5 Relative to C18



Retention on C18 Relative to RP-AmideC16



NOTE: k' ratios not equal to 1.0 mean different retention.

HPLC: Small Molecules

Rediscover Method Development

Different Selectivity is a General and Valuable Characteristic of Functionalized Reversed-Phases

Key to interpreting results

When a color aligns, the selectivity is similar.

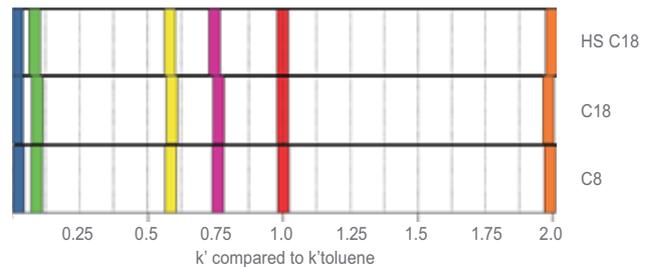
When a color does not align, the selectivity is different.



Mobile phase: 45:55 25 mM Potassium Phosphate (pH 7.0):MeOH (All columns except HS PEG run at 75:25 25 mM Potassium Phosphate (pH 7.0):MeOH).
Flow Rate: 1.0mL/min

Similar Phases (C18's and C8's) - Similar Selectivities

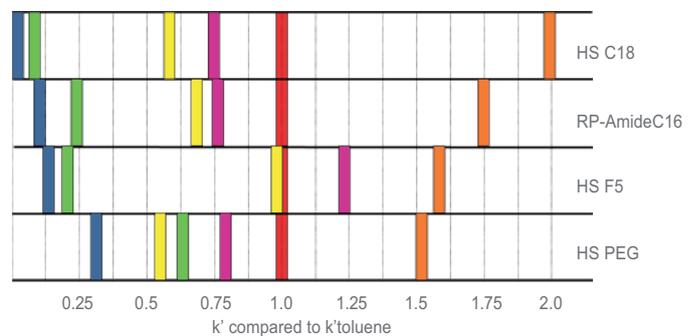
The near-perfect alignment of the colors clearly illustrates the similar selectivities of the C18 and C8 phases tested.



Unique, Functionalized Phases - Different Selectivities

The functional group containing solutes - Aniline, Phenol, N,N-Dimethylaniline (N,N-DMA) and Ethylbenzoate - clearly illustrates the very different selectivities of the functionalized reversed-phases vs. C18. Observe the colors representing solutes containing polar groups dramatically change positions from phase to phase. Also observe the changing hydrophobic selectivity by looking at the Ethylbenzene bar.

Both polar and hydrophobic selectivities are different on the different phases.



Tips for Getting Started: Good Method Development Practices

Tip One: Column Selector Valve

Automated HPLC + Column Selector Valve

- While screening of functionalized reversed-phases can be done with a simple, manual HPLC system, an automated, multi-solvent system with programmable, temperature controlled column selector valve is highly recommended.

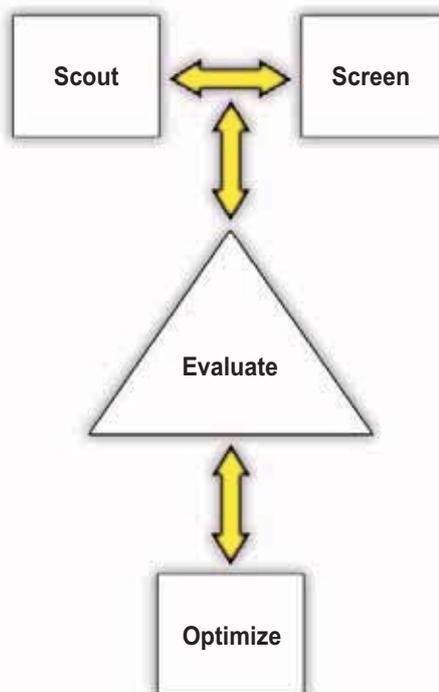
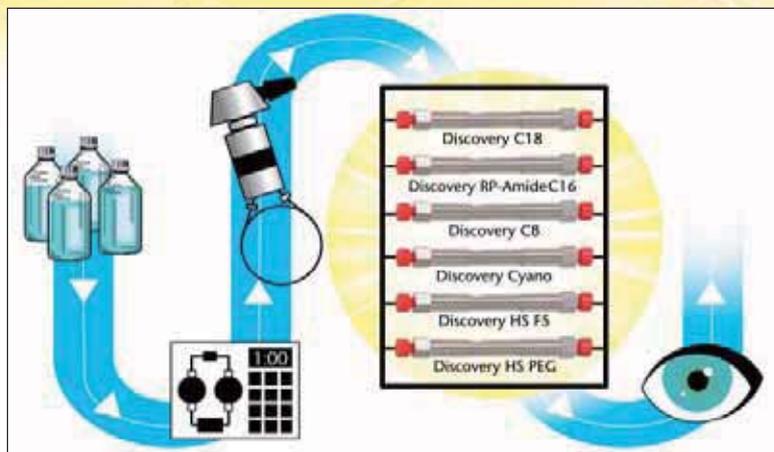
Tip Two: Simple Column Screening

Guidelines for Rapid Screening of Functionalized Reversed-Phases

- Step 1: Scout for "best" mobile phase on C18
- Step 2: Initial screening runs
 - Chromatograph sample on Discovery RP-AmideC16 and F5 using "best" C18 mobile phase
 - Chromatograph sample on PEG using 20% lower organic than "best" C18 mobile phase
- Step 3: Evaluate screening runs
 - Retention OK? If no, adjust % organic and rerun (Note: F5 sometimes requires stronger mobile phase than C18)
- Step 4: Optimize separation on most promising 1 or 2 columns using standard reversed-phase mobile phase adjustment techniques

Tip Three: Always screen several functionalized reversed-phases and a C18.

Tip Four: Optimize your separation on the 1 or 2 most promising phases!



HPLC: Small Molecules

Alkyl Reversed-Phases

Discovery C18, C8, HS C18....

C18 and C8

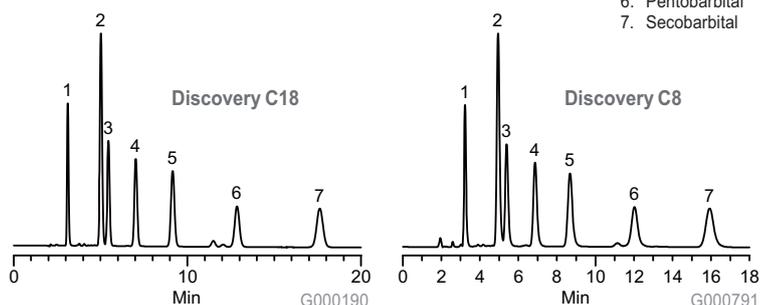
- Excellent performance
- Similar selectivities
- C18 generally more retentive than C8

	Surface Area: (m ² /g)	Pore Size: (Å)	Coverage: (μmoles/m ²)	Endcapped	%C
C8	200	180	3.4	Yes	7.5%
C18	200	180	3.0	Yes	12%
HSC18	300	120	3.8	Yes	20%

Barbiturates

Column: 15cm x 4.6mm columns, 5μm particles
 Mobile Phase: 55:45 Water:MeOH
 Flow Rate: 1.0mL/min
 Det.: UV at 214nm
 Temp.: ambient
 Inj.: 5μL (Discovery C8) or 10μL (Discovery C18)

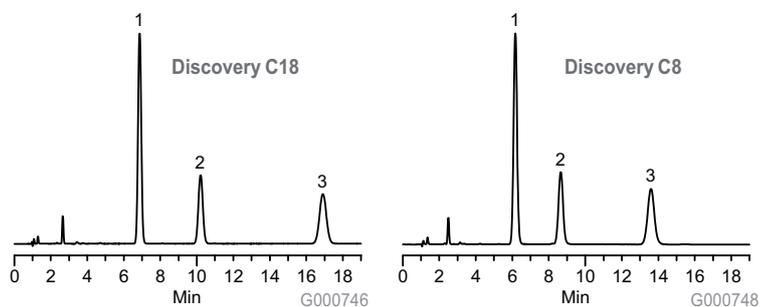
Sample: 1μg/mL of each
 1. Barbitol
 2. Aprobarbital
 3. Phenobarbital
 4. Butabarbital
 5. Mephobarbital
 6. Pentobarbital
 7. Secobarbital



Anticonvulsants

Column: 15cm x 4.6mm columns, 5μm particles
 Mobile Phase: 70:30 Water:CH₃CN
 Flow Rate: 2.0mL/min
 Det.: UV at 254nm
 Temp.: 20°C
 Inj.: 10μL

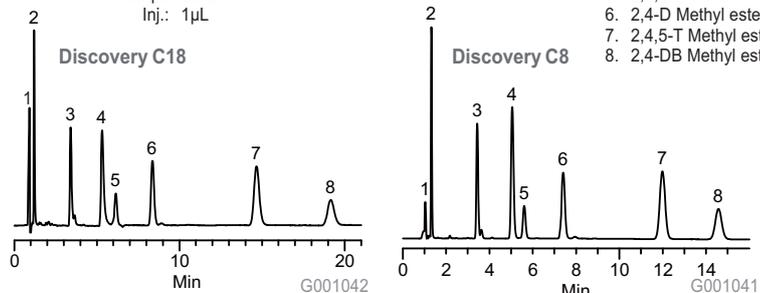
Sample:
 1. Clonazepam
 2. Clorazepate
 3. Diazepam



Alhanoic / Aryloxyalhanoic Acid Using Isocratic Elution

Column: 15cm x 4.6mm columns, 5μm particles
 Mobile Phase: 60:40 25mM Potassium Phosphate (pH 2.3):CH₃CN
 Flow Rate: 2.0mL/min
 Det.: UV at 214nm
 Temp.: 20°C
 Inj.: 1μL

Sample:
 1. Solvent
 2. Dalapon
 3. 2,4-D
 4. 2,4-DB
 5. 2,4,5-T
 6. 2,4-D Methyl ester
 7. 2,4,5-T Methyl ester
 8. 2,4-DB Methyl ester



Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Small Molecules Alkyl Reversed-Phases

...Excellent Chromatography for a Wide Range of Compounds

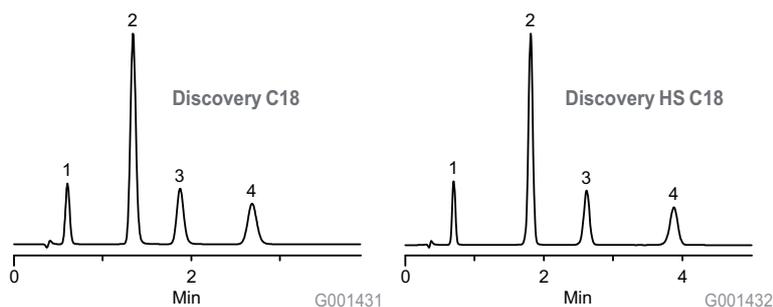
HS C18 and C18

- C18 most general, 1st choice
- Similar selectivities
- HS C18 generally more retentive than C18
- HS C18 is more acidic
 - Mobile phase additive can improve chromatography of bases

Organic Acids

Column: 5cm x 4.6mm columns
Mobile Phase: 60:40 0.1%TFA in Water:MeOH
Flow Rate: 2.0mL/min
Temp.: 20°C
Det.: UV at 254nm
Inj.: 10µL

Sample:
1. Homovanillic acid (0.0625µg/mL)
2. Sorbic acid (0.00625µg/mL)
3. Salicylic acid (0.0625µg/mL)
4. p-Toluic acid (0.00625µg/mL)

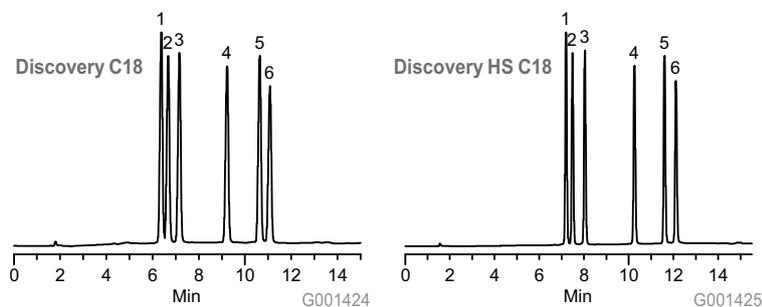


Antibiotics (Fluoroquinolones from Tablets)

Column: 15cm x 4.6mm columns
Mobile Phase: (A) 25 mM Potassium Phosphate (pH 3.0)
(B) CH₃CN
Flow Rate: 1.0mL/min
Temp.: 35°C
Det.: UV at 220nm
Inj.: 10µL

Sample:
1. Levofloxacin
2. Ciprofloxacin
3. Lomefloxacin
4. Sparfloxacin
5. Grepafloxacin
6. Trovafloxacin

Gradient:	Min	%A	%B
	0	90	10
	15	65	35



Conditions for Discovery C18 - no additive

Column: 15cm x 4.6mm columns
5µm particles
Mobile Phase: 45:55 25mM Ammonium Phosphate (pH7.0):CH₃CN
Flow Rate: 1.0mL/min
Temp.: 30°C
Det.: UV at 254nm
Inj.: 10µL
Sample: 100µg/mL of each

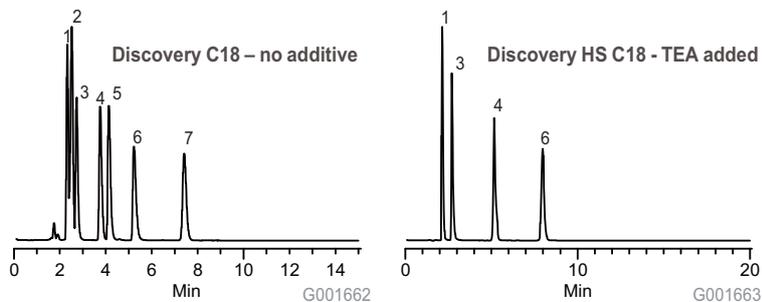
Conditions for Discovery HS C18 - TEA added

Column: 15cm x 4.6mm columns
5µm particles
Mobile Phase: 45:55 25mM TEA phosphate (pH7):CH₃CN
Flow Rate: 1.0mL/min
Temp.: 30°C
Det.: UV at 254nm
Inj.: 10µL
Sample: 100µg/mL of each

...HS C18 is More Acidic...

Sample:
1. Nordoxepin
2. Protriptyline/Desipramine
3. Nortriptyline

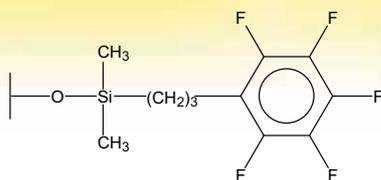
4. Doxepin
5. Imipramine
6. Amitriptyline
7. Trimipramine



HPLC: Small Molecules

Discovery HS F5

Unique Retention and Selectivity Enables Better Separations



- Excellent performance
- Unique selectivity
- Similar retention to C18 (sometimes requires stronger mobile phase strength than C18)

	Surface Area: (m ² /g)	Pore Size: (Å)	Coverage: (μmoles/m ²)	Endcapped	%C
HS F5	300	120	4.0	Yes	12

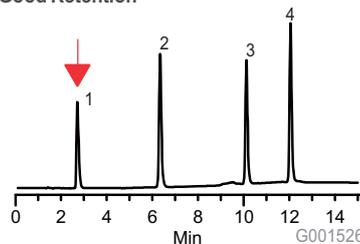
Excellent Retention of Multifunctional Compounds

The Discovery HS F5 shows greater retention, versus C18, of the multifunctional compounds shown in these chromatograms. Compounds that elute too closely to the void volume (peak 1) on C18 columns are sufficiently retained by Discovery HS F5.

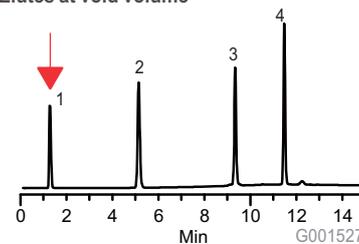
Column: Discovery HS F5 and Conventional C18, 15cm x 4.6mm, 5μm particles
 Mobile Phase: (A) 10mM ammonium acetate, 0.1% formic acid; (B) MeOH
 Flow Rate: 1.5mL/min
 Temp.: 35°C
 Det.: UV at 254nm
 Inj.: 10μL

Gradient:	Min	%A	%B	Sample:
	0	90	10	1. p-Aminophenol (100μg/mL)
	3	90	10	2. Acetaminophen (10μg/mL)
	10	50	50	3. Acetanilide (10μg/mL)
	15	50	50	4. Phenacetin (10μg/mL)

Discovery HS F5:
Good Retention



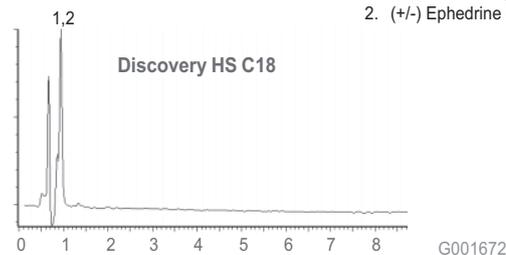
Conventional C18 Phase:
Elutes at void volume



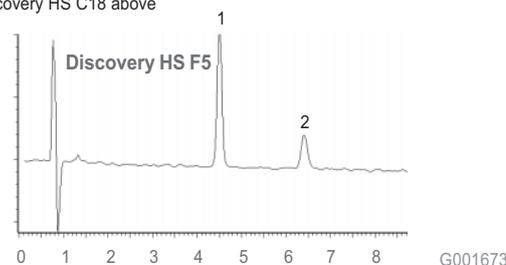
F5 Provides Excellent Separation - Solutes Are Not Retained on C18

Column: Discovery HS C18, 15cm x 4.6mm, 5μm particles
 Mobile Phase: 30:70 10mM Ammonium Acetate (pH=6.98): CH₃CN
 Flow Rate: 2.0mL/min
 Temp.: 35°C
 Det.: Photodiode Array
 Inj.: 5μL

Sample:
1. Methcathinone (100μg/mL)
2. (+/-) Ephedrine (200μg/mL)



Column: Discovery HS F5, 15cm x 4.6mm
 Conditions same as Discovery HS C18 above



HPLC: Small Molecules Discovery HS F5

F5, a unique, functionalized reversed-phase uncovers a trace impurity missed by C18. "Pure" Quinidine was assayed on C18 under a variety of mobile phase conditions. Conditions C and D produced a single peak suggesting the Quinidine was pure. The peak resulting from condition B might be showing a partially resolved front shoulder. A quick screen of % organic was unable to resolve the possible impurity.

On F5 (chromatogram A) the impurity is clearly resolved. During method development a quick screen using unique, functionalized reversed-phases such as F5 greatly increases the chances of finding trace impurities early, before they can cause big problems.

Helpful Hint

Routinely screen separations on several, complimentary functionalized reversed-phases (e.g. F5, PEG, RP-AmideC16) early in method development.

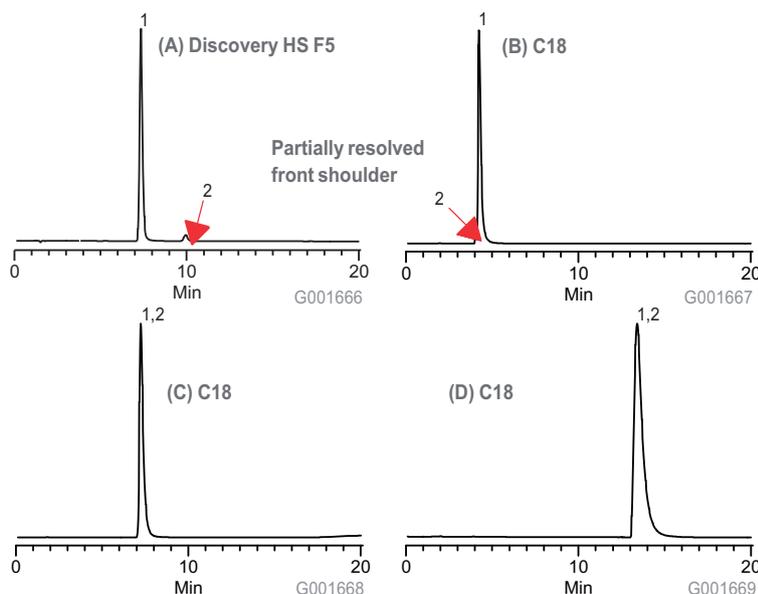
At lower organic, Discovery HS F5 exhibits reversed-phase behavior. At higher organic it exhibits normal phase behavior. At high % organic retention actually increases as % organic increases for some analytes. Benefits include:

- Improve LC/MS detection by using higher % organic mobile phase.
- Use mobile phase selectivity to develop valuable, different separations at high % organic

F5 Resolves Trace Impurity in Quinidine – C18 Does Not!

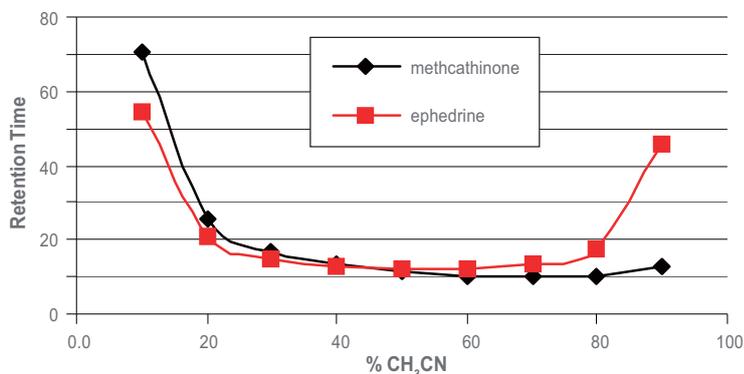
Column: Discovery HS F5 and Conventional C18, 15cm x 4.6mm, 5 μ m particles
Mobile Phase: 25 mM Ammonium Phosphate (pH 7.0):CH₃CN.
Varying Ratios: (A) 35:65, (B) 70:30, (C) 76:24, (D) 80:20
Flow Rate: 1.0mL/min
Temp.: 30°C
Det.: UV at 235nm
Inj.: 10 μ L

Sample:
1. Quinidine (50 μ g/mL)
2. Impurity



F5 Exhibits "U" Shaped Retention Creating Interesting Possibilities

F5 - Retention vs. % Organic



HPLC: Small Molecules

Discovery RP-Amide C16

Unique Retention and Selectivity Enables Better Separations

- Excellent performance
- Unique selectivity
- Similar retention to C18 (typically requires similar mobile phase strength as C18)

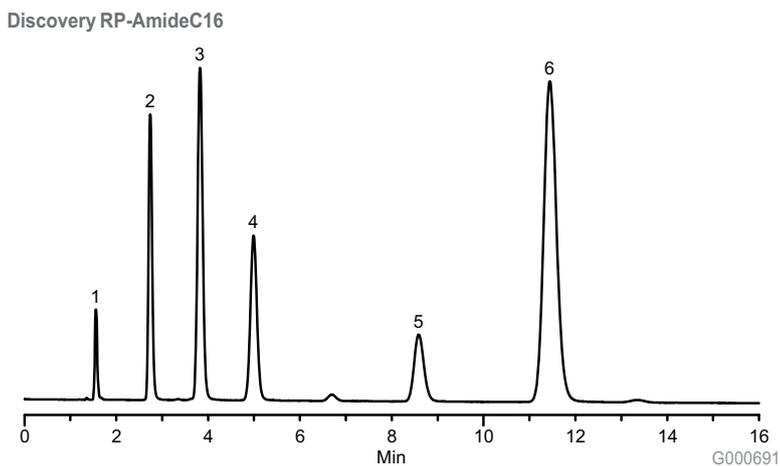
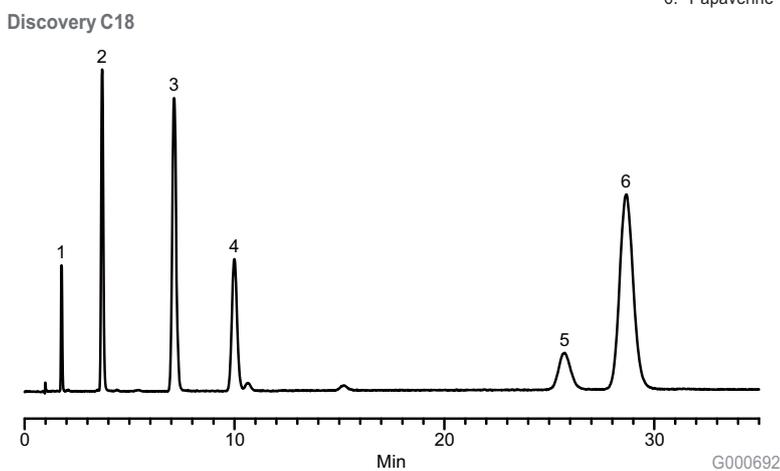
Discovery RP-AmideC16 Gives Better Resolution and Faster Analysis

- faster analysis from lower hydrophobicity
- better peak spacing (RP-AmideC16)
- better resolution of small impurities (RP-AmideC16)

Column: 15cm x 4.6mm, 5µm particles
 Mobile Phase: 80:20 25mM Potassium Phosphate (pH 3.0):MeOH
 Flow Rate: 2.0mL/min
 Temp.: 35°C
 Det.: UV at 254nm
 Inj.: 10µL

- Sample:**
1. Codeine
 2. Strychnine
 3. Quinidine
 4. Quinine
 5. Noscapine
 6. Papaverine

	Surface Area: (m ² /g)	Pore Size: (Å)	Coverage: (µmoles/m ²)	Endcapped	%C
RP-AmideC16	200	180	2.6	Yes	11%



Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Small Molecules

Discovery RP-Amide C16

Faster analysis and different selectivity.

Antitussive/Antihistamine/Antipyretic Mix

Dramatic differences in peak order and run time are demonstrated by different Discovery reversed-phase stationary phases.

Column: 15cm x 4.6mm, 5 μ m particles
 Mobile Phase: (A) 25 mM Potassium Phosphate (pH 2.3)
 (B) CH₃CN
 Flow Rate: 2.0mL/min
 Temp.: ambient
 Det.: UV at 214nm
 Inj.: 10 μ L

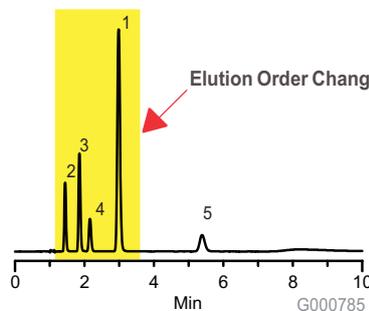
Sample: (1 μ g/mL of each)

1. Acetaminophen
2. Doxylamine
3. Pseudoephedrine
4. Codeine
5. Chlorpheniramine

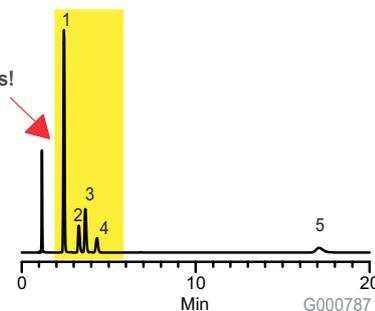
Gradient:

Min	%A	%B
0	90	10
2	90	10
4	70	30
8	70	30
10	50	50

Discovery RP-AmideC16



Discovery C18



But... different selectivity is not always better!

Here C18 provides a better separation.

Barbiturates

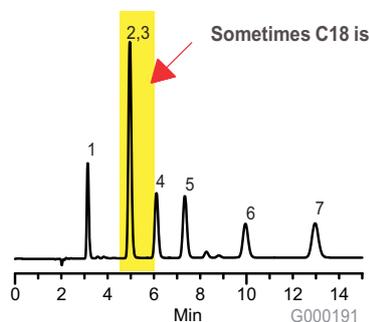
Different Discovery column chemistries show different separations of barbiturates under identical conditions.

Column: 15cm x 4.6mm columns, 5 μ m particles
 Mobile Phase: 55:45 Water:MeOH
 Flow Rate: 1.0mL/min
 Temp.: ambient
 Det.: UV at 214nm
 Inj.: 10 μ L

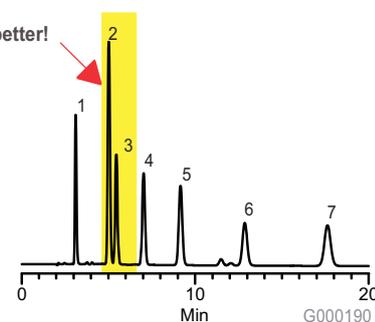
Sample: (1 μ g/mL of each)

1. Barbitol
2. Aprobarbital
3. Phenobarbital
4. Butabarbital
5. Mephobarbital
6. Pentobarbital
7. Secobarbital

Discovery RP-AmideC16



Discovery C18



Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

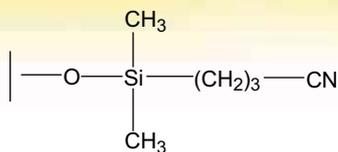
Liquid Chromatography

SUPELCO

HPLC: Small Molecules

Discovery Cyano

Unique Retention and Selectivity Enables Better Separations



- Excellent performance
- Unique selectivity
- Significantly less retentive than C18 (therefore typically requires greater % H₂O mobile phase)

	Surface Area: (m ² /g)	Pore Size: (Å)	Coverage: (μmoles/m ²)	Endcapped	%C
Cyano	200	180	3.5	Yes	4.5

Faster Analysis - Eliminate Wasted Time

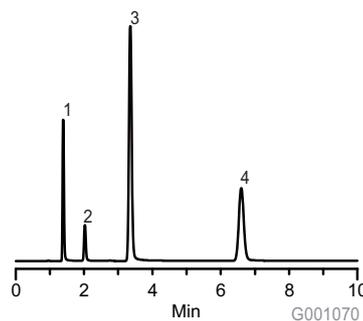
Urea Pesticides Using Isocratic Elution

Column: 15cm x 4.6mm columns, 5μm particles
 Mobile Phase: 60:40 Water:CH₃CN
 Flow Rate: 2.0mL/min
 Temp.: 20°C,
 Det.: UV at 214nm
 Inj.: 1μL

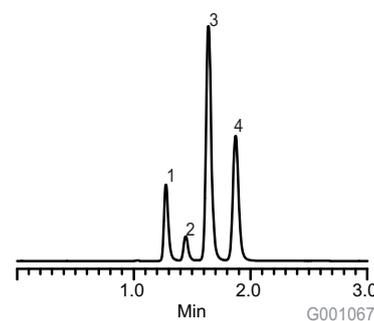
Sample:

1. Fenuron
2. Monuron
3. Diuron
4. Linuron

Discovery C18



Discovery Cyano



Faster Analysis - Different Selectivity

Organophosphorous Pesticides Using Isocratic Elution

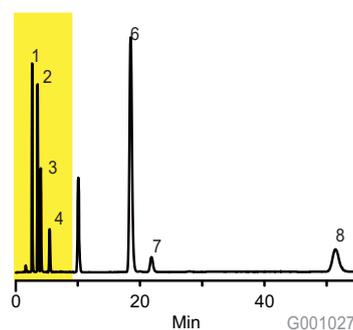
Column: Discovery C18, 15cm x 4.6mm, 5μm particles
 Mobile Phase: 30:70 Water:MeOH
 Flow Rate: 1.0mL/min
 Temp.: 20°C
 Det.: UV at 214nm
 Inj.: 1μL

Sample:

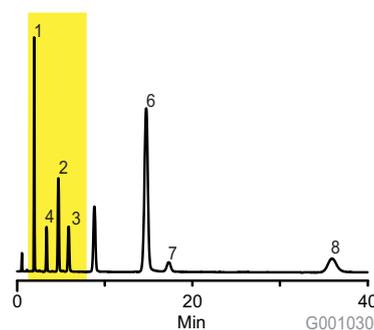
1. Dichlorvos
2. Guthion
3. Methyl parathion
4. Ethoprophos
5. Disulfoton
6. Fenchlorvos
7. Chlorpyrifos
8. Prothiophos

Column: Discovery Cyano, 15cm x 4.6mm, 5μm particles
 Mobile Phase: 75:25 Water:CH₃CN
 Flow Rate: 2.0mL/min
 Temp.: 20°C
 Det.: UV at 214nm
 Inj.: 1μL

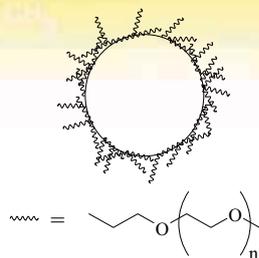
Discovery C18



Discovery Cyano



Unique Retention and Selectivity Enables Better Separations



- Excellent performance
- Unique selectivity
- Significantly less retentive than C18 (therefore typically requires greater % H_2O mobile phase)

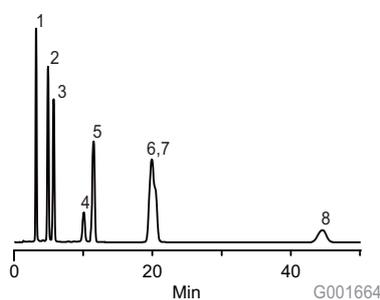
	Surface Area: (m^2/g)	Pore Size: (\AA)	Coverage: ($\mu\text{moles}/\text{m}^2$)	Encapped	%C
HS PEG	300	120	3.8	No	12

Flavones Demonstrate Dramatic Selectivity Differences Between HS PEG and C18

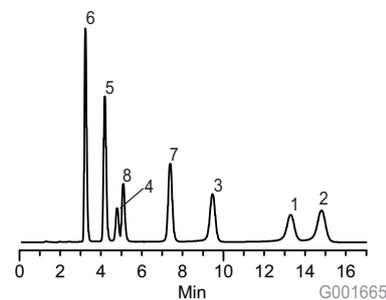
Column: 15cm x 4.6mm columns, 5 μm particles
 Mobile Phase: 45:55 0.1% Formic Acid in Water :
 0.1% Formic Acid in MeOH
 Flow Rate: 1.0mL/min
 Temp.: 30°C,
 Det.: UV at 254nm
 Inj.: 10 μL

Sample: 50 $\mu\text{g}/\text{mL}$ of each
 1. Myricetin
 2. Quercetin
 3. Luteolin
 4. Baicalein
 5. 7-Hydroxyflavone
 6. Flavone
 7. Chrysin
 8. 5-Hydroxyflavone

Discovery HS C18



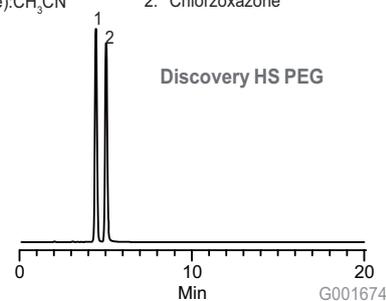
Discovery HS PEG



Chlorzoxazone - Excellent Separation on PEG; Excessive Retention and Resolution on HS C18

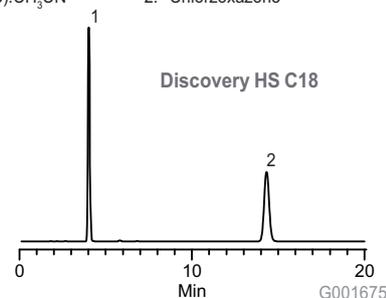
Column: 15cm x 4.6mm columns, 5 μm particles
 Mobile Phase: 70:30 20mM Acetic Acid in Water
 (pH 4.5 with Ammonium Hydroxide): CH_3CN
 Flow Rate: 1.0mL/min
 Temp.: 30°C,
 Det.: UV at 285 nm
 Inj.: 10 μL

Sample: 100 $\mu\text{g}/\text{mL}$ of each
 1. 6-Hydroxychlorzoxazone
 2. Chlorzoxazone



Column: 15cm x 4.6mm columns, 5 μm particles
 Mobile Phase: 75:25 20mM Acetic Acid in Water
 (pH 4.5 with Ammonium Hydroxide): CH_3CN
 Flow Rate: 1.0mL/min
 Temp.: 30°C,
 Det.: UV at 285 nm
 Inj.: 10 μL

Sample: 100 $\mu\text{g}/\text{mL}$ of each
 1. 6-Hydroxychlorzoxazone
 2. Chlorzoxazone



HPLC: Small Molecules

Discovery C18 and Discovery HS C18 Columns

Discovery C18

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY C18 (180Å, 12% CARBON)			
5µm	5 x 2.1	50494721	
5µm	10 x 2.1	569220-U	
5µm	12.5 x 2.1	569229-U	
5µm	15 x 2.1	50495521	
5µm	5 x 3.0	504947-30	
5µm	10 x 3.0	569221-U	
5µm	12.5 x 3.0	569230-U	
5µm	15 x 3.0	504955-30	
5µm	25 x 3.0	504971-30	
5µm	5 x 4.0	504947-40	
5µm	10 x 4.0	569222-U	
5µm	12.5 x 4.0	569231-U	
5µm	15 x 4.0	504955-40	
5µm	25 x 4.0	504971-40	
5µm	5 x 4.6	504947	
5µm	10 x 4.6	569223-U	
5µm	12.5 x 4.6	569232-U	
5µm	15 x 4.6	504955	
5µm	25 x 4.6	504971	
5µm	25 x 10.0	569224-U	
5µm	25 x 21.2	569226-U	

DISCOVERY C18 VALIDATION PACKS*

5µm	5 x 2.1	55700-U21
5µm	10 x 2.1	569800-U
5µm	15 x 2.1	55702-U21
5µm	5 x 4.6	55700-U
5µm	10 x 4.6	569801-U
5µm	15 x 4.6	55702-U
5µm	25 x 4.6	55704-U

DISCOVERY C18 SUPELGUARD CARTRIDGES

5µm	2 x 2.1 (2/pk)	505188
5µm	2 x 2.1 kit **	505161
5µm	2 x 3.0 (2/pk)	59576-U
5µm	2 x 3.0 kit **	59575-U
5µm	2 x 4.0 (2/pk)	505137
5µm	2 x 4.0 kit **	505129

* Packs include 3 columns, each from a different lot of bonded phase.

** Kits include one cartridge, a stand-alone holder, a piece of tubing, 2 nuts and ferrules.

DISCOVERY C18 PROPERTIES:

Bonded Phase:	Octadecylsilane, endcapped
Silica:	Spherical, high purity (Fe <20; NA <7; Ca <7; Ti <1; Al <1; Mg <1ppm)
Particle Size:	5µm
Pore Size:	180Å
Surface Area:	200m ² /g
%C:	~12%
Coverage:	~3µmoles/m ²

Discovery HS C18

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY HS C18 (120Å, 20% CARBON)			
3µm	5 x 2.1	569253-U	
3µm	7.5 x 2.1	569254-U	
3µm	15 x 2.1	569255-U	
3µm	5 x 4.6	569250-U	
3µm	7.5 x 4.6	569251-U	
3µm	15 x 4.6	569252-U	
5µm	5 x 2.1	568500-U	
5µm	10 x 2.1	568501-U	
5µm	15 x 2.1	568502-U	
5µm	25 x 2.1	568503-U	
5µm	5 x 4.0	568510-U	
5µm	10 x 4.0	568511-U	
5µm	15 x 4.0	568512-U	
5µm	25 x 4.0	568513-U	
5µm	5 x 4.6	568520-U	
5µm	10 x 4.6	568521-U	
5µm	15 x 4.6	568522-U	
5µm	25 x 4.6	568523-U	
5µm	5 x 10.0	568530-U	
5µm	10 x 10.0	568531-U	
5µm	15 x 10.0	568532-U	
5µm	25 x 10.0	568533-U	
5µm	5 x 21.2	568540-U	
5µm	10 x 21.2	568541-U	
5µm	15 x 21.2	568542-U	
5µm	25 x 21.2	568543-U	
10µm	5 x 10.0	568630-U	
10µm	10 x 10.0	568631-U	
10µm	15 x 10.0	568632-U	
10µm	25 x 10.0	568633-U	
10µm	5 x 21.2	568640-U	
10µm	10 x 21.2	568641-U	
10µm	15 x 21.2	568642-U	
10µm	25 x 21.2	568643-U	

DISCOVERY HS C18 SUPELGUARD CARTRIDGES

3µm	2 x 2.1 (2/pk)	569276-U
3µm	2 x 2.1 kit **	569277-U
3µm	2 x 4.0 (2/pk)	569274-U
3µm	2 x 4.0 kit **	569275-U
5µm	2 x 2.1 (2/pk)	568570-U
5µm	2 x 2.1 kit **	568571-U
5µm	2 x 4.0 (2/pk)	568572-U
5µm	2 x 4.0 kit **	568573-U
5µm	1 x 10.0	568574-U
10µm	1 x 10.0	568674-U

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

DISCOVERY HS C18 PROPERTIES:

Bonded Phase:	Octadecylsilane, endcapped
Silica:	Spherical, high purity (<10 ppm metals)
Particle Size:	3, 5, 10µm
Pore Size:	120Å
Surface Area:	300m ² /g
%C:	~20%
Coverage:	~3.8µmoles/m ²

HPLC: Small Molecules

Discovery RP-AmideC16 and Discovery HS F5 Columns

Discovery RP-AmideC16

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY RP-AMIDEC16 (180Å, 11% CARBON)			
5µm	5 x 2.1	50500521	
5µm	10 x 2.1	569320-U	
5µm	12.5 x 2.1	569329-U	
5µm	15 x 2.1	50501321	
5µm	5 x 3.0	505005-30	
5µm	10 x 3.0	569321-U	
5µm	12.5 x 3.0	569330-U	
5µm	15 x 3.0	505013-30	
5µm	25 x 3.0	505064-30	
5µm	5 x 4.0	505005-40	
5µm	10 x 4.0	569322-U	
5µm	12.5 x 4.0	569331-U	
5µm	15 x 4.0	505013-40	
5µm	25 x 4.0	505064-40	
5µm	5 x 4.6	505005	
5µm	10 x 4.6	569323-U	
5µm	12.5 x 4.6	569332-U	
5µm	15 x 4.6	505013	
5µm	25 x 4.6	505064	
5µm	25 x 10.0	569324-U	
5µm	25 x 21.2	569326-U	

DISCOVERY RP-AMIDEC16 VALIDATION PACKS *

5µm	5 x 2.1	55705-U21
5µm	10 x 2.1	569802-U
5µm	15 x 2.1	55707-U21
5µm	5 x 4.6	55705-U
5µm	10 x 4.6	569803-U
5µm	15 x 4.6	55707-U
5µm	25 x 4.6	55709-U

DISCOVERY RP-AMIDEC16 SUPELGUARD CARTRIDGES

5µm	2 x 2.1 (2/pk)	505110
5µm	2 x 2.1 kit **	505102
5µm	2 x 3.0 (2/pk)	59578-U
5µm	2 x 3.0 kit **	59577-U
5µm	2 x 4.0 (2/pk)	505099
5µm	2 x 4.0 kit **	505080

* Packs include 3 columns, each from a different lot of bonded phase.

** Kits include one cartridge, a stand-alone holder, a piece of tubing, 2 nuts and ferrules.

DISCOVERY RP-AMIDEC16 PROPERTIES:

Bonded Phase:	Palmitamidopropylsilane, endcapped
Silica:	Spherical, high purity (Fe <20; NA <7; Ca <7; Ti <1; Al <1; Mg <1ppm)
Particle Size:	5µm
Pore Size:	180Å
Surface Area:	200m ² /g
%C:	~11%
Coverage:	~2.6µmoles/m ²

Discovery HS F5 New!

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY HS F5 (120Å, 12% CARBON)			
3µm	5 x 2.1	567500-U	
3µm	10 x 2.1	567502-U	
3µm	15 x 2.1	567503-U	
3µm	5 x 4.0	567530-U	
3µm	10 x 4.0	567531-U	
3µm	15 x 4.0	567532-U	
3µm	5 x 4.6	567504-U	
3µm	10 x 4.6	567506-U	
3µm	15 x 4.6	567507-U	
5µm	5 x 2.1	567508-U	
5µm	10 x 2.1	567510-U	
5µm	15 x 2.1	567511-U	
5µm	25 x 2.1	567512-U	
5µm	5 x 4.0	567533-U	
5µm	10 x 4.0	567534-U	
5µm	15 x 4.0	567535-U	
5µm	25 x 4.0	567536-U	
5µm	5 x 4.6	567513-U	
5µm	10 x 4.6	567515-U	
5µm	15 x 4.6	567516-U	
5µm	25 x 4.6	567517-U	
5µm	5 x 10.0	567518-U	
5µm	10 x 10.0	567537-U	
5µm	15 x 10.0	567519-U	
5µm	25 x 10.0	567520-U	
5µm	5 x 21.2	567521-U	
5µm	10 x 21.2	567539-U	
5µm	15 x 21.2	567522-U	
5µm	25 x 21.2	567523-U	
10µm	5 x 10.0	567524-U	
10µm	10 x 10.0	567538-U	
10µm	15 x 10.0	567525-U	
10µm	25 x 10.0	567526-U	
10µm	5 x 21.2	567527-U	
10µm	10 x 21.2	567540-U	
10µm	15 x 21.2	567528-U	
10µm	25 x 21.2	567529-U	

DISCOVERY HS F5 SUPELGUARD CARTRIDGES

3µm	2 x 2.1 (2/pk)	567570-U
3µm	2 x 2.1 kit **	567571-U
3µm	2 x 4.0 (2/pk)	567572-U
3µm	2 x 4.0 kit **	567573-U
5µm	2 x 2.1 (2/pk)	567574-U
5µm	2 x 2.1 kit **	567575-U
5µm	2 x 4.0 (2/pk)	567576-U
5µm	2 x 4.0 kit **	567577-U
5µm	1 x 10.0	567578-U
10µm	1 x 10.0	567580-U

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

DISCOVERY HS F5 PROPERTIES:

Bonded Phase:	Pentafluorophenylpropylsilane, endcapped
Silica:	Spherical, high purity (<10 ppm metals)
Particle Size:	3, 5, 10µm
Pore Size:	120Å
Surface Area:	300m ² /g
%C:	~12%
Coverage:	~4µmoles/m ²

HPLC: Small Molecules

Discovery HS PEG and Discovery C8 Columns

Discovery HS PEG



PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY HS PEG (120Å, 12% CARBON)			
3µm	5 x 2.1	567400-U	
3µm	10 x 2.1	567402-U	
3µm	15 x 2.1	567403-U	
3µm	5 x 4.0	567430-U	
3µm	10 x 4.0	567431-U	
3µm	15 x 4.0	567432-U	
3µm	5 x 4.6	567404-U	
3µm	10 x 4.6	567406-U	
3µm	15 x 4.6	567407-U	
5µm	5 x 2.1	567408-U	
5µm	10 x 2.1	567410-U	
5µm	15 x 2.1	567411-U	
5µm	25 x 2.1	567412-U	
5µm	5 x 4.0	567433-U	
5µm	10 x 4.0	567434-U	
5µm	15 x 4.0	567435-U	
5µm	25 x 4.0	567436-U	
5µm	5 x 4.6	567413-U	
5µm	10 x 4.6	567415-U	
5µm	15 x 4.6	567416-U	
5µm	25 x 4.6	567417-U	
5µm	5 x 10.0	567418-U	
5µm	10 x 10.0	567437-U	
5µm	15 x 10.0	567419-U	
5µm	25 x 10.0	567420-U	
5µm	5 x 21.2	567421-U	
5µm	10 x 21.2	567439-U	
5µm	15 x 21.2	567422-U	
5µm	25 x 21.2	567423-U	
10µm	5 x 10.0	567424-U	
10µm	10 x 10.0	567438-U	
10µm	15 x 10.0	567425-U	
10µm	25 x 10.0	567426-U	
10µm	5 x 21.2	567427-U	
10µm	10 x 21.2	567440-U	
10µm	15 x 21.2	567428-U	
10µm	25 x 21.2	567429-U	

DISCOVERY HS PEG SUPELGUARD CARTRIDGES

3µm	2 x 2.1 (2/pk)	567470-U
3µm	2 x 2.1 kit **	567471-U
3µm	2 x 4.0 (2/pk)	567472-U
3µm	2 x 4.0 kit **	567473-U
5µm	2 x 2.1 (2/pk)	567474-U
5µm	2 x 2.1 kit **	567475-U
5µm	2 x 4.0 (2/pk)	567476-U
5µm	2 x 4.0 kit **	567477-U
5µm	1 x 10.0	567478-U
10µm	1 x 10.0	567480-U

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

DISCOVERY HS PEG PROPERTIES:

Bonded Phase:	Polyethyleneglycol, endcapped
Silica:	Spherical, high purity (<10 ppm metals)
Particle Size:	3, 5, 10µm
Pore Size:	120Å
Surface Area:	300m ² /g
%C:	~12%
Coverage:	~3.8µmoles/m ²

Discovery C8

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY C8 (180Å, 7.5% CARBON)			
5µm	5 x 2.1	59352-U21	
5µm	10 x 2.1	569420-U	
5µm	12.5 x 2.1	569424-U	
5µm	15 x 2.1	59353-U21	
5µm	5 x 3.0	59352-U30	
5µm	10 x 3.0	569421-U	
5µm	12.5 x 3.0	569425-U	
5µm	15 x 3.0	59353-U30	
5µm	25 x 3.0	59354-U30	
5µm	5 x 4.0	59352-U40	
5µm	10 x 4.0	569422-U	
5µm	12.5 x 4.0	569426-U	
5µm	15 x 4.0	59353-U40	
5µm	25 x 4.0	59354-U40	
5µm	5 x 4.6	59352-U	
5µm	10 x 4.6	569423-U	
5µm	12.5 x 4.6	569427-U	
5µm	15 x 4.6	59353-U	
5µm	25 x 4.6	59354-U	

DISCOVERY C8 VALIDATION PACKS *

5µm	5 x 2.1	55710-U21
5µm	10 x 2.1	569804-U
5µm	15 x 2.1	55712-U21
5µm	5 x 4.6	55710-U
5µm	10 x 4.6	569805-U
5µm	15 x 4.6	55712-U
5µm	25 x 4.6	55714-U

DISCOVERY C8 SUPELGUARD CARTRIDGES

5µm	2 x 2.1 (2/pk)	59588-U
5µm	2 x 2.1 kit**	59587-U
5µm	2 x 3.0 (2/pk)	59580-U
5µm	2 x 3.0 kit**	59579-U
5µm	2 x 4.0 (2/pk)	59590-U
5µm	2 x 4.0 kit**	59589-U

* Packs include 3 columns, each from a different lot of bonded phase.

** Kits include one cartridge, a stand-alone holder, a piece of tubing, 2 nuts and ferrules.

DISCOVERY C8 PROPERTIES:

Bonded Phase:	Octylsilane, endcapped
Silica:	Spherical, high purity (Fe <20; NA <7; Ca <7; Ti <1; Al <1; Mg <1ppm)
Particle Size:	5µm
Pore Size:	180Å
Surface Area:	200m ² /g
%C:	~7.5%
Coverage:	~3.4µmoles/m ²

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Small Molecules

Discovery Cyano Columns, Discovery Selectivity Packs

Discovery Cyano

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY CYANO (180Å, 4.5% CARBON)			
5µm	5 x 2.1	59355-U21	
5µm	10 x 2.1	569521-U	
5µm	12.5 x 2.1	569524-U	
5µm	15 x 2.1	59356-U21	
5µm	5 x 3.0	59355-U30	
5µm	10 x 3.0	569522-U	
5µm	12.5 x 3.0	569525-U	
5µm	15 x 3.0	59356-U30	
5µm	25 x 3.0	59357-U30	
5µm	5 x 4.0	59355-U40	
5µm	10 x 4.0	569523-U	
5µm	12.5 x 4.0	569526-U	
5µm	15 x 4.0	59356-U40	
5µm	25 x 4.0	59357-U40	
5µm	5 x 4.6	59355-U	
5µm	10 x 4.6	569520-U	
5µm	12.5 x 4.6	569527-U	
5µm	15 x 4.6	59356-U	
5µm	25 x 4.6	59357-U	

DISCOVERY CYANO VALIDATION PACKS*

5µm	5 x 2.1	55715-U21
5µm	10 x 2.1	569806-U
5µm	15 x 2.1	55717-U21
5µm	5 x 4.6	55715-U
5µm	10 x 4.6	569807-U
5µm	15 x 4.6	55717-U
5µm	25 x 4.6	55719-U

DISCOVERY CYANO SUPELGUARD CARTRIDGES

5µm	2 x 2.1 (2/pk)	59584-U
5µm	2 x 2.1 kit**	59583-U
5µm	2 x 3.0 (2/pk)	569571-U
5µm	2 x 3.0 kit**	569570-U
5µm	2 x 4.0 (2/pk)	59586-U
5µm	2 x 4.0 kit**	59585-U

* Packs include 3 columns, each from a different lot of bonded phase.

** Kits include one cartridge, a stand-alone holder, a piece of tubing, 2 nuts and ferrules.

DISCOVERY CYANO PROPERTIES:

Bonded Phase:	Cyanopropylsilane, endcapped
Silica:	Spherical, high purity (Fe <20; NA <7; Ca <7; Ti <1; Al <1; Mg <1ppm)
Particle Size:	5µm
Pore Size:	180Å
Surface Area:	200m ² /g
%C:	~4.5%
Coverage:	~3.5µmoles/m ²

Discovery Selectivity Packs

You can conveniently order the four Discovery column chemistries – RP-AmideC16, C18, C8, and Cyano – in your choice of column dimensions, in a single kit. Quickly evaluate mobile phases on all four columns to find the optimum combination of chemistries for your separation. The Discovery HPLC Column Selectivity Pack gives you a powerful tool for rapid, efficient, simple pharmaceutical method development.

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
5µm	5 x 2.1	55720-U21	
5µm	10 x 2.1	569853-U	
5µm	15 x 2.1	55722-U21	
5µm	5 x 3.0	55720-U30	
5µm	10 x 3.0	569852-U	
5µm	15 x 3.0	55722-U30	
5µm	25 x 3.0	55724-U30	
5µm	5 x 4.0	55720-U40	
5µm	10 x 4.0	569851-U	
5µm	15 x 4.0	55722-U40	
5µm	25 x 4.0	55724-U40	
5µm	5 x 4.6	55720-U	
5µm	10 x 4.6	569850-U	
5µm	15 x 4.6	55722-U	
5µm	25 x 4.6	55724-U	

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

Liquid
Chromatography

SUPELCO

HPLC: Small Molecules

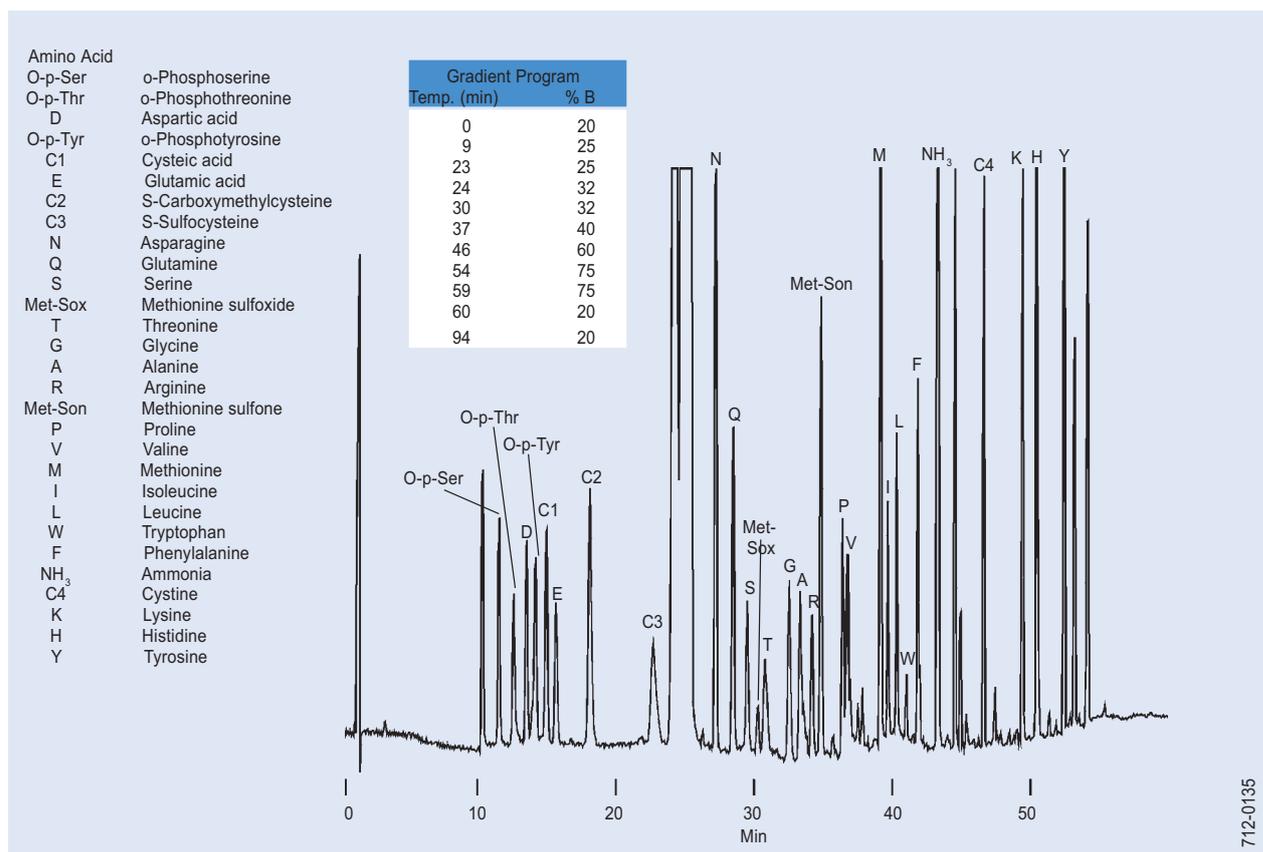
Special Purpose SUPELCOSIL Columns

Special Purpose Columns: Amino Acids

SUPELCOSIL LC-DABS columns feature a specially treated and tested octadecylsilane bonded phase, for reversed-phase separations of precolumn derivatized dabsyl amino acids. More than 30 amino acids and ammonia can be separated in less than one hour.

PROPERTIES

Particles:	spherical silica, 3µm
Pore Size:	120Å
Bonded Phase:	octadecylsilane
Surface Area:	170m ² /g
Pore Volume:	0.6mL/g
pH Range:	2 - 7.5



Dabsyl Amino Acids

Column: SUPELCOSIL LC-DABS
15cm x 4.6mm ID, 3µm particles
Cat. No.: 59137
Mobile Phase: A = 25mM KH₂PO₄, pH7.0
B = acetonitrile:methanol (70:30)
Flow Rate: 1.5mL/min
Det.: VIS, 436nm
Inj.: 5µL, approx. 50pM each derivative

RELATED INFORMATION

Literature References
Stocchi, V., et al., Anal. Biochem. 178: 107-117 (1989).
Stocchi, V., et al., Amino Acids 3: 303-309 (1992).
References not available from Supelco.

PARTICLE SIZE	LENGTH X ID		CAT. NO.	PRICE
	(cm X mm)			
SUPELCOSIL LC-DABS (120Å, 12.3% CARBON)				
3µm	15 x 4.6		59137	
SUPELCOSIL LC-18-T Supelguard Cartridges (use for LC-DABS)				
5µm	2 x 4.0 (2/pk)		59621	
5µm	2 x 4.0 kit **		59620	

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

Special Purpose SUPELCOSIL Columns

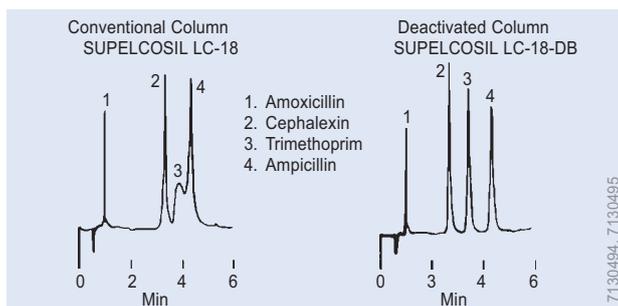
Special Purpose Columns: Basic Columns

SUPELCOSIL LC-18-DB and SUPELCOSIL LC-8-DB phases are deactivated (DB) for basic compounds. These columns provide shorter retention, better peak shape, and higher efficiency for organic bases than can be obtained on conventional reversed-phase columns. DB columns have become standards in the industry for analyses of "difficult" basic compounds, particularly in the pharmaceutical industry. An example of the performance of the LC-18-DB column versus that of a conventional C18 column is shown. The deactivated column allows the use of a simpler mobile phase to elute trimethoprim with good peak shape. To obtain similar results from the LC-18 column would require the addition of competing base to the mobile phase

PROPERTIES

Silica: Spherical
 Particle size: 3µm and 5µm
 Pore size: 120Å
 Pore Volume: 0.6mL/g
 Surface Area: 170m²/g
 pH Range: 2-7

Improved Peak Symmetry for Antibacterials



Column: 3.3cm x 4.6mm, 3µm
 Cat. No.: 58977 (LC-18), 58978 (LC-18-DB)
 Mobile Phase: methanol:50mM KH₂PO₄, pH 3.0 (23:77)
 Flow Rate: 1mL/min
 Det.: UV, 254nm
 Inj.: 10µL, 15, 5, 5 and 106µg/mL

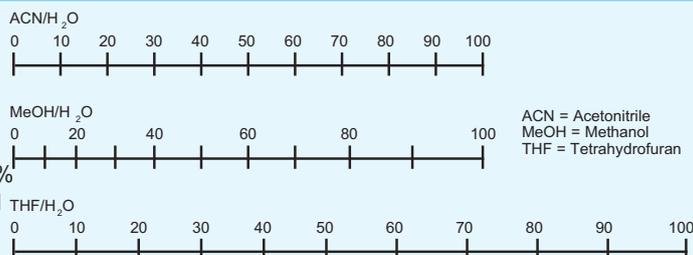
PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCOSIL LC-18-DB (120Å, 11.0% CARBON)			
3µm	25 x 2.1	57943	
3µm	3.3 x 3.0	58978C30	
3µm	7.5 x 3.0	58992C30	
3µm	15 x 3.0	58993C30	
3µm	15 x 4.0	58993C40	
3µm	3.3 x 4.6	58978	
3µm	7.5 x 4.6	58992	
3µm	15 x 4.6	58993	
5µm	30 x 1.0	57984	
5µm	25 x 2.1	57940	
5µm	5 x 3.0	58345C30	
5µm	10 x 3.0	59208C30	
5µm	15 x 3.0	58348C30	
5µm	25 x 3.0	58355C30	
5µm	15 x 4.0	58348C40	
5µm	25 x 4.0	58355C40	
5µm	30 x 4.0	59164	
5µm	5 x 4.6	58345	
5µm	10 x 4.6	59208	
5µm	15 x 4.6	58348	
5µm	25 x 4.6	58355-U	
5µm	25 x 10.0	58358	
SUPELCOSIL LC-18-DB Supelguard Cartridges			
5µm	2 x 2.1 (2/pk)	59617	
5µm	2 x 2.1 kit **	59616	
5µm	2 x 3.0 (2/pk)	59565C30	
5µm	2 x 4.0 (2/pk)	59565	
5µm	2 x 4.0 kit **	59555	

SUPELCOSIL LC-8-DB (120Å, 6.0% CARBON)			
3µm	7.5 x 3.0	58990C30	
3µm	15 x 3.0	58991C30	
3µm	15 x 4.0	58991C40	
3µm	3.3 x 4.6	58976	
3µm	7.5 x 4.6	58990-U	
3µm	15 x 4.6	58991	
5µm	25 x 2.1	57933	
5µm	15 x 3.0	58347C30	
5µm	15 x 4.0	58347C40	
5µm	25 x 4.0	58354C40	
5µm	5 x 4.6	58344	
5µm	15 x 4.6	58347	
5µm	25 x 4.6	58354	
5µm	25 x 10.0	58357	
SUPELCOSIL LC-8-DB Supelguard Cartridges			
5µm	2 x 2.1 (2/pk)	59619	
5µm	2 x 2.1 kit **	59618	
5µm	2 x 4.0 (2/pk)	59563	
5µm	2 x 4.0 kit **	59553	

HELPFUL HINTS

Relative Strengths for Different Solvents

The graph provides for the interconversion of reversed-phase mobile phases having the same strength. Vertical lines in this figure intersect mobile phases having the same strength. For example, 40% Acetonitrile has the same strength as 50% Methanol or 30% THF.



** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

Special Purpose SUPELCO SIL Columns

Special Purpose Columns: Carbohydrates

Within the different classes of sugars, chemical and physical properties vary only slightly. HPLC separations of carbohydrates depend on differences in conformation, configuration, and column type. Because of this complexity, no single HPLC column or method is capable of separating every carbohydrate. To choose the best column for your carbohydrate analysis, consult Table 1 (below), the Retention Time Index (next page), and the Applications section of this catalog. For more information, request Bulletin 887H (HPLC Carbohydrate Column Selection Guide).

SUPELCOGEL K columns separate raffinose, sucrose, glucose, fructose, and betaine, a trimethylammonium zwitterionic compound found in beet and cane sugars and widely distributed in other plants.

The lead-form resin in SUPELCOGEL Pb columns provides the highest resolution and best selectivity for monosaccharides. SUPELCOGEL Pb columns provide excellent separation of xylose, galactose, and mannose, which are not completely resolved on calcium-form resin columns.

SUPELCOGEL Ca columns separate monosaccharides and sugar alcohols. Di-, tri-, and oligosaccharides are separated by class. A frequent application for this column is the separation of sugars in high fructose corn syrup (HFCS).

SUPELCOGEL C-610H and H columns are ideal for separating carbohydrates, alcohols, and organic acids present in the same sample: wines and other fermentation products, fruit juices, biological samples, etc.

SUPELCOGEL C-611 columns contain a unique ion exchange resin containing two divalent cations, rather than one. This provides different selectivities for separating monosaccharides

and sugar alcohols. As with resins containing a single cation, di-, tri-, and oligosaccharides are separated by class. Galactose and mannose are well separated.

SUPELCOGEL Ag columns provide rapid separations of oligosaccharides. Glycerol and ethanol are well resolved.

Silica-based SUPELCO SIL LC-NH₂ columns separate monosaccharides, disaccharides, and some trisaccharides, with elution generally in order of increasing molecular weight. Retention decreases as the proportion of water:acetonitrile in the mobile phase is increased. For more information, request T397126 (Sugars on SUPELCO SIL LC-NH₂ Columns).

SUPELCOGEL CARBOHYDRATE COLUMN CHARACTERISTICS

Particles:	sulfonated polystyrene/divinylbenzene, spherical, 9µm
Counter Ion:	varies (see Table 1)
pH Range:	1-13
Organic Compatibility:	<10% in mobile phase
Maximum Temperature:	varies (see Table 1)
Maximum Flow Rate:	7.8mm ID columns: 1.5mL/min 4.6mm ID columns: 0.4mL/min
Maximum Pressure:	1000psi (70 bar)

SUPELCO SIL LC-NH₂ COLUMN CHARACTERISTICS

Particles:	spherical silica, 5µm
Bonded Phase:	aminopropylsilyl
pH Range:	2-7.5
Organic Compatibility:	no limits (avoid aldehydes and ketones)
Maximum Flow Rate:	2mL/min (4.6mm ID columns)
Maximum Pressure:	6000psi (420 bar)

Table 1. Carbohydrate Column Applications and Mobile Phases

COLUMN	APPLICATION ¹	FORM	TYPICAL MOBILE PHASE	MAX. TEMP. (°C)
SUPELCOGEL K	beet sugar, cane sugar, molasses, corn syrup	potassium	10mM H ₂ KPO ₄	90
SUPELCOGEL Pb	monosaccharides, xylose/galactose/mannose	lead	deionized water	90
SUPELCOGEL Ca	high fructose corn syrup, monosaccharides, sugar alcohols, oligosaccharides	calcium	deionized water	90
SUPELCOGEL C-610H	organic acids	hydrogen	0.1% H ₂ SO ₄ or H ₃ PO ₄	60
SUPELCOGEL H	organic acids	hydrogen	0.1% H ₂ SO ₄ or H ₃ PO ₄	90
SUPELCOGEL C-611	mono-, di-, and trisaccharides, galactose/mannose	2 divalent cations	10N NaOH	85
SUPELCOGEL Ag1	beer, dark corn syrup	silver	deionized water	90
SUPELCOGEL Ag2	oligosaccharides, glycerol/ethanol, corn syrup, hydrolyzed starch	silver	deionized water	90
SUPELCO SIL LC-NH ₂	mono-, di-, some trisaccharides	aminopropyl silica	75% CH ₃ CN in water	70

¹ See Applications pages.

HPLC: Small Molecules

Special Purpose SUPELCOSIL Columns

Table 2. Retention Time Index for Carbohydrate Columns

Cat. No.:	SUPELCOGEL COLUMNS									SUPELCOSIL LC-NH ₂ 58338
	Ca 59305-U	C-610H 59320-U	H 59304-U	H 59346	Pb 59343	C-611 59310-U	K 59342	Ag2 59315		
Dimensions (mm):	300 x 7.8	300 x 7.8	300 x 7.8	250 x 4.6	300 x 7.8	300 x 7.8	300 x 7.8	300 x 7.8	300 x 7.8	250 x 4.6
Temp.:	80°C	30°C	30°C	30°C	85°C	60°C	85°C	85°C	85°C	ambient
Mobile Phase:	DH ₂ O	0.1% H ₃ PO ₄	0.1% H ₃ PO ₄	0.1% H ₃ PO ₄	DH ₂ O	10 ⁻⁴ N NaOH	15mM K ₂ HPO ₄	DH ₂ O	ACN:DH ₂ O (3:1)	
Flow Rate (mL/min):	0.5	0.5	0.5	0.17	0.5	0.5	0.5	0.5	0.5	1.0
Det.:	refractive index									
Compound Retention Times (min)										
Arabinose	15.3	13.9	14.3	13.8	19.2	19.6	16.8	17.1	7.5	
Arabitol	19.8	14.1	14.9	14.3	32.3	22.8	13.5	16.0	7.2	
Betaine	ND	ND	ND	ND	NR	ND	13.0	ND	ND	
Dulcitol	22.3	13.4	14.2	13.7	43.4	25.7	12.9	15.9	9.0	
Erythritol	17.7	15.0	15.6	14.8	24.5	20.2	14.0	16.1	5.9	
Ethanol	19.4	25.6	ND	ND	ND	21.0	ND	18.4	NR	
Fructose	14.9	13.1	13.3	12.9	20.8	20.7	15.2	16.0	8.3	
Galactose	13.4	12.9	13.0	12.6	17.6	17.6	15.1	15.8	10.3	
Glucose	12.0	12.1	11.9	11.7	14.9	15.8	14.0	14.6	9.8	
Glycerol	18.7	16.8	17.6	16.6	23.8	20.9	15.2	17.1	NR	
Inositol	14.9	12.6	12.7	12.4	24.5	20.1	15.7	17.4	ND	
Isomaltose	9.6	10.3	ND	ND	ND	13.8	ND	11.6	19.4	
Isomaltotriose	8.5	9.5	ND	ND	ND	12.6	ND	9.8	NR	
Lactitol	ND	ND	11.1	11.0	26.5	ND	10.6	ND	ND	
Lactose	10.2	10.8	10.2	10.2	13.5	14.3	10.9	11.8	19.5	
Maltitol	13.6	11.0	10.7	10.7	23.8	17.7	10.2	15.0	15.5	
Maltoheptaose	7.5	8.8	7.6	7.9	9.2	11.6	7.2	7.3	NR	
Maltohexaose	7.7	8.9	7.7	8.1	9.7	12.0	7.4	7.6	NR	
Maltopentaose	7.9	9.1	7.9	8.2	10.5	12.6	7.8	8.1	NR	
Maltose	9.8	10.5	9.9	9.9	13.0	14.2	10.7	11.5	17.4	
Maltotetraose	8.3	9.3	8.2	8.5	11.2	13.2	8.4	8.8	NR	
Maltotriose	8.8	9.7	8.8	9.0	12.0	13.6	9.2	9.8	31.0	
Mannitol	19.2	13.2	13.7	13.2	32.5	22.1	12.6	15.2	9.2	
Mannose	13.7	12.8	12.9	12.5	19.8	18.9	15.6	15.9	9.1	
Melezitose	8.7	9.7	8.8	9.0	10.8	12.4	8.6	9.3	24.5	
Psicose	22.5	13.4	14.5	13.9	36.5	32.9	15.5	17.2	6.6	
Raffinose	8.7	9.7	8.7	8.9	11.2	12.6	8.7	9.6	29.7	
Ribitol	16.7	13.7	14.2	13.6	25.1	19.5	13.1	15.3	ND	
Ribose	24.3	14.2	15.8	15.0	40.7	34.6	17.7	19.1	6.0	
Sorbitol	23.4	13.4	14.4	13.9	46.9	28.3	13.3	16.3	9.0	
Stachyose	8.1	9.3	8.1	8.4	10.4	11.9	7.9	8.5	67.3	
Sucrose	9.8	10.6	9.9	9.9	12.2	13.6	10.1	11.2	14.0	
Xylitol	23.3	14.4	15.7	15.0	42.1	28.0	14.2	17.1	7.3	
Xylose	13.2	12.8	12.8	12.6	16.1	17.2	15.3	15.6	6.8	

NR - not recommended
ND - no data available
For optimal separations, allow at least 1 minute between compounds.

For resins used in processing sugars and foods, refer to the low pressure LC media section.

SUPELCOGEL and SUPELCOSIL Carbohydrate Columns and Guard Columns

COLUMN	LENGTH (cm)	ID (mm)	CAT. NO.	PRICE	SUPELGUARD GUARD COLUMN	CAT. NO.	PRICE
SUPELCOGEL K	30	7.8	59342		K*	59344	
SUPELCOGEL Pb	30	7.8	59343		Pb*	59345	
SUPELCOGEL Ca	30	7.8	59305-U		Ca*	59306-U	
SUPELCOGEL C-610H	30	7.8	59320-U		H*	59319	
SUPELCOGEL H	30	7.8	59304-U		H*	59319	
SUPELCOGEL H	25	4.6	59346		H*	59319	
SUPELCOGEL C-611	30	7.8	59310-U		Ca*	59306-U	
SUPELCOGEL Ag1	30	7.8	59318-U		Ag1*	59317-U	
SUPELCOGEL Ag2	30	7.8	59315		Ag2*	59316	
SUPELCOSIL LC-NH ₂	25	4.6	58338		LC-NH ₂ (kit)** LC-NH ₂ (2 cartridges)	59558 59568	

* 5cm x 4.6mm guard column, does not include tubing, nuts or ferrules.

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

Special Purpose SUPELCOSIL Columns

COLUMN CHARACTERISTICS

Particles: 5 μ m spherical silica
 Bonded Phase: aminopropylsilyl
 Pore Size: 120Å
 Surface Area: 170m²/g
 Pore Volume: 0.6mL/g
 pH Range: 2 - 7.5

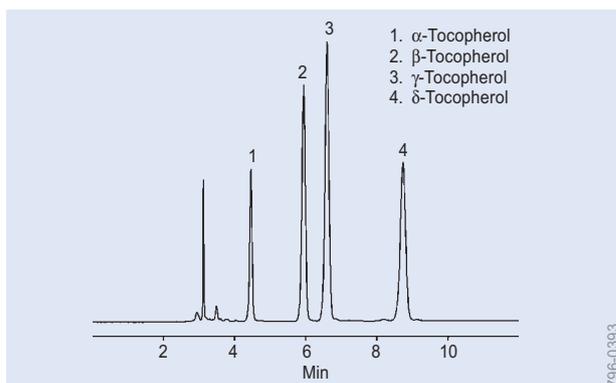


Figure A. Tocopherols

Column: SUPELCOSIL LC-NH₂-NP, 25cm x 4.6mm ID, 5 μ m particles
 Cat. No.: 59132
 Mobile Phase: hexane:ethyl acetate, 70:30
 Flow Rate: 1.0mL/min
 Temp.: 30°C
 Det.: UV, 295nm
 Inj.: 10 μ L hexane, 1.0mg/mL each analyte

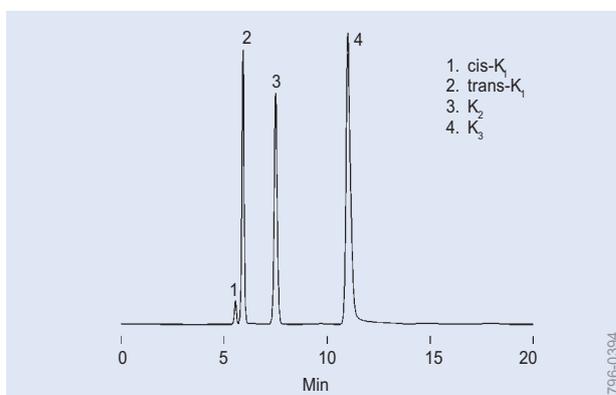


Figure B. Vitamin K Isomers

Column: SUPELCOSIL LC-NH₂-NP, 25cm x 4.6mm ID, 5 μ m particles
 Cat. No.: 59132
 Mobile Phase: hexane:ethyl acetate, 99:1
 Flow Rate: 1.5mL/min
 Temp.: 30°C
 Det.: UV, 254nm
 Inj.: 10 μ L hexane, 0.3mg/mL each analyte

Special Purpose Columns: Dedicated Normal Phase

SUPELCOSIL LC-NH₂-NP is an amino phase dedicated to normal phase chromatography. By employing special bonding technology, and avoiding water in manufacturing and testing the column, we have dramatically reduced the retention variation that is characteristic of normal phase chromatography.

SUPELCOSIL LC-NH₂-NP columns:

- show stable retention in normal phase separations
- are less sensitive to small or varying amounts of water in the mobile phase, relative to unmodified silica
- provide excellent separations of fat-soluble vitamins

Normal phase chromatography is especially useful when the analytes are not water soluble – for example, the fat-soluble vitamins A, D, E, and K. Figure A shows a separation of tocopherols (vitamin E) on a SUPELCOSIL LC-NH₂-NP column. The various isomers of vitamin K are separated in Figure B.

These columns should be used with non-aqueous mobile phases only.

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCOSIL LC-NH ₂ -NP (120Å, 3.0% CARBON)			
5 μ m	25 x 4.6	59132	
SUPELCOSIL LC-NH ₂ -NP Supelguard Cartridges			
5 μ m	2 x 4.0 (2/pk)	59516	
5 μ m	2 x 4.0 kit **	59515	

HELPFUL HINTS

Reversed-phase versus Normal Phase
 Reversed-phase is characterized by strong interactions between analytes and the polar mobile phase. Interactions between analytes and the nonpolar stationary phase are weak. Mobile phases typically consist of water / organic solvent combinations. Reversed-phase columns include: Amide-C16, C18, C8, Phenyl, C5, Pentafluorinated Phenyl (F5), Cyano, C1, ODP-50, and TPR-100.

Normal phase is characterized by strong interactions between analytes and the polar stationary phase. Interactions between analytes and the nonpolar mobile phase are weak. Mobile phases consist of organic solvents. Normal phase columns include: Cyano, NH₂, and Silica.

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

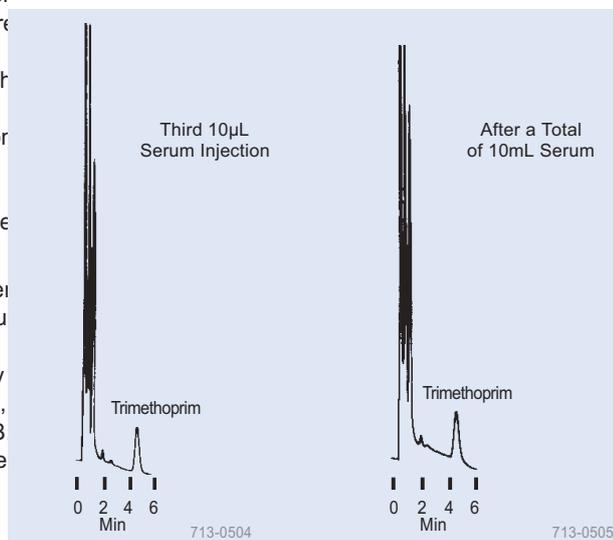
Special Purpose SUPELCOSIL Columns

Special Purpose Columns: Direct Serum Injection

The Hisep column uses a shielded hydrophobic phase (SHP) for its mechanism of separation. The silica-based material is covered with a thin polymer consisting of hydrophobic regions in a hydrophilic network. Small analytes, such as drugs, penetrate the hydrophilic network and are retained by the hydrophobic moieties. The hydrophilic network shields protein molecules from contact with the surface and the hydrophobic groups, and thus these molecules are not retained. Direct injection of biological samples eliminates time-consuming cleanup steps and increases analytical accuracy.

Excellent column stability and an ability to exclude proteins over a wide pH range set Hisep columns apart from other direct serum injection columns and techniques. Stability is demonstrated in Figure A – no significant change is seen in the chromatography of trimethoprim after injecting 10mL of serum, in 10 μ L increments, onto a Hisep column. A low pH application is shown in Figure B. The profile of the excluded serum peak will change with pH due to differing protonation states of the serum proteins.

Figure A. Hisep Columns Perform Consistently for Many Injections



Column: Hisep, 15cm x 4.6mm ID, 5 μ m particles

Cat. No.: 58935

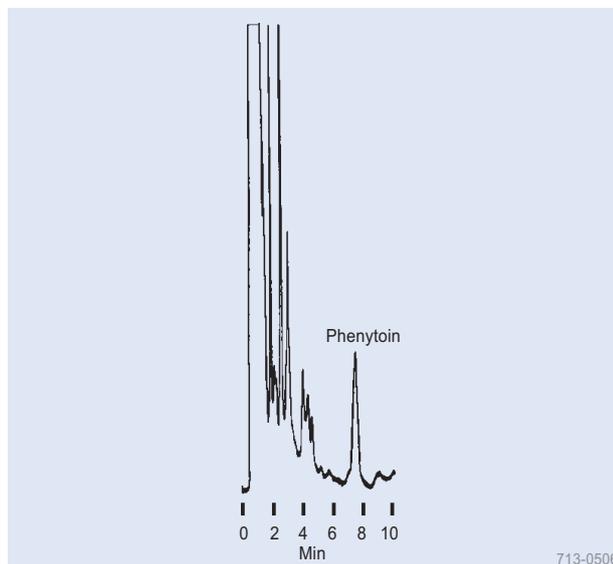
Mobile Phase: acetonitrile:180mM ammonium acetate (15:85), pH 7

Flow Rate: 2mL/min

Det.: UV, 254nm

Inj.: 10 μ L spiked serum (25 μ g/mL trimethoprim)

Figure B. Hisep Columns Are Compatible with Low pH Mobile Phases



Column: Hisep, 15cm x 4.6mm ID, 5 μ m particles

Cat. No.: 58935

Mobile Phase: acetonitrile:180mM ammonium acetate (pH 4.6)

Flow Rate: 2mL/min

Det.: UV, 254nm

Inj.: 25 μ L phenytoin-spiked serum

PROPERTIES

Particles:	spherical silica, 5 μ m
Pore Size:	120Å
Surface Area:	170m ² /g
Pore Volume:	0.6mL/g
pH range:	2-7.5

SUPELCOSIL Hisep

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCOSIL HISEP (120Å)			
5 μ m	25 x 2.1	57932	
5 μ m	5 x 4.6	59143	
5 μ m	15 x 4.6	58935	
5 μ m	25 x 4.6	58919	
SUPELCOSIL Hisep Supelguard Cartridges			
5 μ m	2 x 4.0 (2/pk)	59640-U	
5 μ m	2 x 4.0 kit **	59639	

RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No.	Subject
T397145	Hisep columns for drugs

Literature References

Feibush, B., C. Santasania. *J. Chromatogr.*54: 441-449 (1991).
 Gisch, D.J., B.T. Hunter, B. Feibush. *J. Chromatogr.*433: 264-268(1988).
 Wong, S.H.Y., L.A. Bretts, A.C. Larson. *J. Liq. Chromatogr.*11: 2039-2049 (1988).

References not available from Supelco.

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

Special Purpose SUPELCOSIL Columns

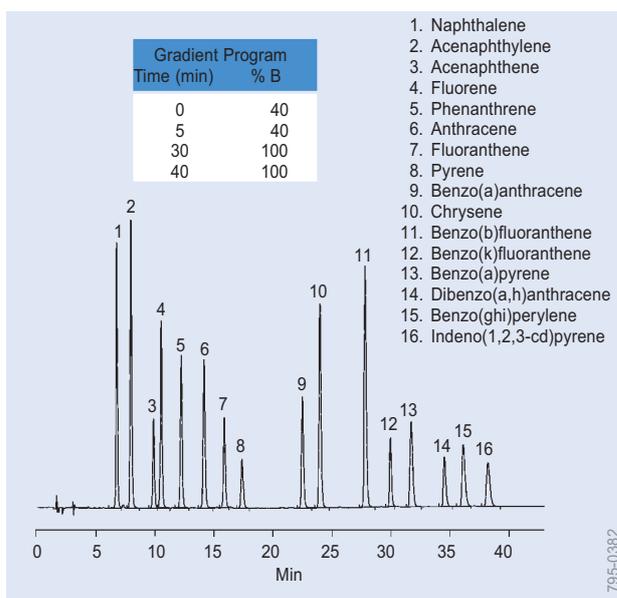


Figure A. Optimum PAH Resolution: 5µm Particles

Column: SUPELCOSIL LC-PAH, 25cm x 4.6mm ID, 5µm particles
 Cat. No.: 58229
 Mobile Phase: A = water, B = acetonitrile
 Flow Rate: 1.5mL/min
 Det.: UV, 254nm
 Inj.: 3µL LC-PAH Test Mix (Cat. No. 48743), diluted 1:10 with acetonitrile

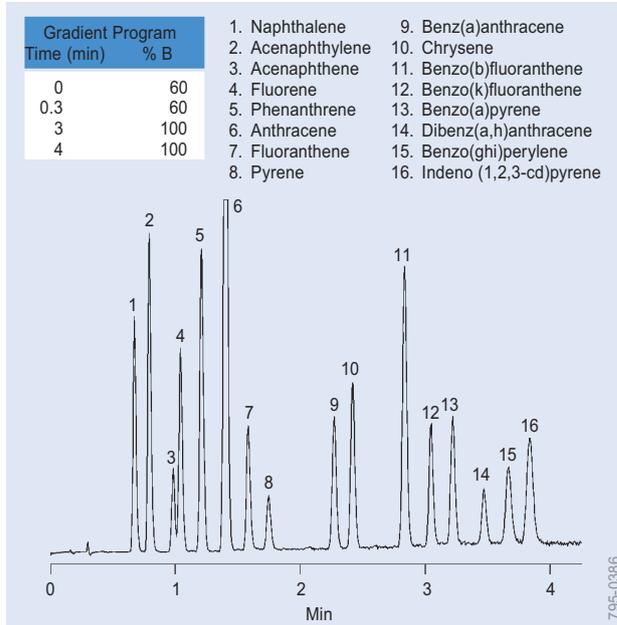


Figure B. Rapid Analyses: 3µm Particles

Column: SUPELCOSIL LC-PAH, 5cm x 4.6mm ID, 3µm particles
 Cat. No.: 59133
 Mobile Phase: A = water, B = acetonitrile
 Flow Rate: 3.0mL/min
 Det.: UV, 254nm

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

Special Purpose Columns: Polyaromatic Hydrocarbons

SUPELCOSIL LC-PAH columns were designed specifically for analyses of the priority pollutant PAHs listed in US EPA Method 610 (Figure A). 2.1mm and 3.0mm columns save solvent and improve sensitivity when sample mass is limited. 3µm columns provide extremely rapid, highly efficient analyses (Figure B), while retaining the durability of porous silicas. They are excellent and economical substitutes for 1.5µm nonporous silicas.

COLUMN CHARACTERISTICS

Particles: spherical silica, 3µm, 5µm
 Pore Size: 120Å
 Bonded Phase: octadecylsilane
 Surface Area: 170m²/g
 Pore Volume: 0.6mL/g
 pH Range: 2 - 7.5

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCOSIL LC-PAH (120Å)			
3µm	5 x 3.0	59133C30	
3µm	10 x 3.0	59134C30	
3µm	5 x 4.6	59133	
3µm	10 x 4.6	59134	
5µm	25 x 2.1	57945	
5µm	15 x 3.0	58318C30	
5µm	15 x 4.6	58318	
5µm	25 x 4.6	58229	
SUPELCOSIL LC-18 Supelguard Cartridges (use for LC-PAH)			
5µm	2 x 2.1 (2/pk)	59613	
5µm	2 x 2.1 kit **	59612	
5µm	2 x 3.0 (2/pk)	59564C30	
5µm	2 x 4.0 (2/pk)	59564	
5µm	2 x 4.0 kit **	59554	

HELPFUL HINTS

Reversed-phase versus Normal Phase
 Reversed-phase is characterized by strong interactions between analytes and the polar mobile phase. Interactions between analytes and the nonpolar stationary phase are weak. Mobile phases typically consist of water / organic solvent combinations. Reversed-phase columns include: Amide-C16, C18, C8, Phenyl, C5, Pentafluorinated Phenyl (F5), Cyano, C1, ODP-50, and TPR-100.

Normal phase is characterized by strong interactions between analytes and the polar stationary phase. Interactions between analytes and the nonpolar mobile phase are weak. Mobile phases consist of organic solvents. Normal phase columns include: Cyano, NH₂, and Silica.

HPLC: Small Molecules

Special Purpose SUPELCOGEL Columns

Special Purpose Columns: Nucleosides

SUPELCOSIL LC-18-S columns are designed for reliable separations of deoxyribonucleosides and ribonucleosides. Each column is tested to ensure performance.

PROPERTIES

Silica:	Spherical
Particle Size:	5µm
Pore Size:	120Å
Bonded Phase:	octadecylsilane
Surface Area:	170m ² /g
Pore Volume:	0.6mL/g
pH Range:	2-7.5

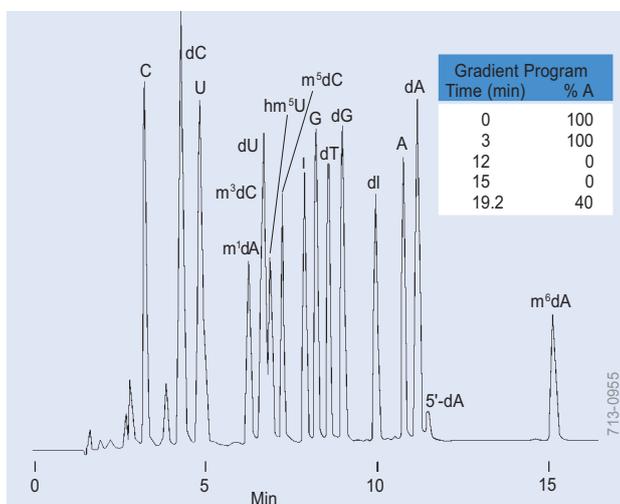


Figure provided by Dr. C. W. Gehrke and K.C. Kuo, University of Missouri-Columbia, Experimental Station Chemical Laboratories, Columbia, MO USA

Deoxyribonucleosides and Ribonucleosides on a SUPELCOSIL LC-18-S Column

Column: SUPELCOSIL LC-18-S, 15cm x 4.6mm ID, 5µm particles
 Cat. No.: 58931
 Mobile Phase: 50mM K₂HPO₄/KH₂PO₄ (pH 4.0): methanol
 A = 97.5:2.5, B = 80:20
 Flow Rate: 1.0mL/min
 Temp.: 30°C
 Det.: UV, 254nm
 Inj.: nucleoside standards in water

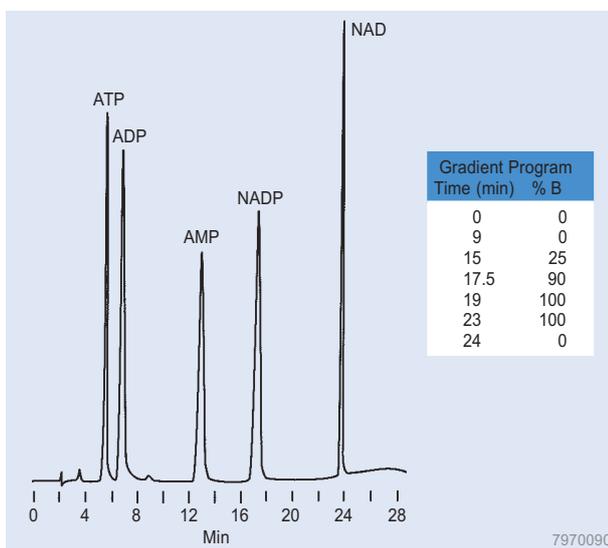
PARTICLE SIZE	LENGTH X ID		CAT. NO.	PRICE
	(cm X mm)			
SUPELCOSIL LC-18-S (120Å, 11.0% CARBON)				
5µm	30 x 1.0		57920	
5µm	25 x 2.1		57939	
5µm	15 x 3.0		58931C30	
5µm	15 x 4.6		58931	
5µm	25 x 4.6		58928-U	
SUPELCOSIL LC-18-S Supelguard Cartridges				
5µm	2 x 4.0 (2/pk)		59630	
5µm	2 x 4.0 kit **		59629	

Special Purpose Columns: Nucleotides

SUPELCOSIL LC-18-T columns feature an octadecylsilane bonded phase and a special surface treatment, for efficient separations of nucleotides. Each batch of packing material is tested to ensure good peak shape for a representative nucleotide, adenosine diphosphate (ADP). Chromatography of other compounds that exhibit metal chelating properties also can be improved by using this phase.

PROPERTIES

Silica:	Spherical
Particle Size:	3µm, 5µm
Pore Size:	120Å
Bonded Phase:	octadecylsilane
Surface Area:	170m ² /g
Pore Volume:	0.6mL/g
pH Range:	2-7.5



Nucleotides on a SUPELCOSIL LC-18-T Column

Column: SUPELCOSIL LC-18-T, 25cm x 4.6mm ID (5µm particles)
 Cat. No.: 58971
 Mobile Phase: A = 0.1M KH₂PO₄, pH 6
 B = A:methanol, 90:10
 gradient program shown on figure
 Flow Rate: 1.3mL/min
 Det.: UV, 254nm

PARTICLE SIZE	LENGTH X ID		CAT. NO.	PRICE
	(cm X mm)			
SUPELCOSIL LC-18-T (120Å, 12.3% CARBON)				
3µm	15 x 3.0		58970C30	
3µm	15 x 4.6		58970-U	
5µm	25 x 3.0		58971C30	
5µm	25 x 4.6		58971	
SUPELCOSIL LC-18-T Supelguard Cartridges				
5µm	2 x 3.0 (2/pk)		59621C30	
5µm	2 x 4.0 (2/pk)		59621	
5µm	2 x 4.0 kit **		59620	

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

Special Purpose SUPELCOGEL Columns

Special Purpose Columns: Organic Acids

SUPELCOGEL C-610H columns are prepared specifically for analyses of organic acids. Acetic, propionic, butyric, formic, malic, citric, succinic, lactic, and other acids are easily separated on these columns, using a simple isocratic mobile phase and minimal sample preparation. Retention times for many acids are shown in the table.

Separation is based on ion exclusion – the analytes selectively partition between the resin phase and the external aqueous phase. Analyses are best performed at low pH (0.1% H_3PO_4 often is used as the mobile phase); the same mobile phase conditions can be applied to a wide variety of sample matrices.

COLUMN CHARACTERISTICS

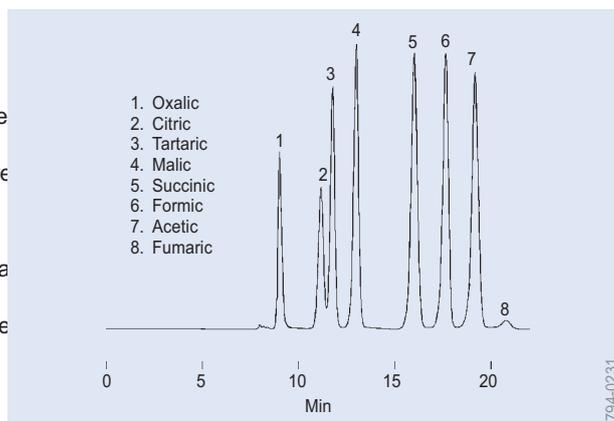
Particles:	sulfonated polystyrene/divinylbenzene, spherical, 9 μ m
Counter Ion:	H ⁺
pH Range:	1-13
Organic Compatibility:	<20% in mobile phase
Maximum Flow Rate:	1.5mL/min
Maximum Pressure:	1000psi (70 bar)

SUPELCOGEL H columns have the same particle composition, retention mechanism, performance, sensitivity, and applications as SUPELCOGEL C-610H columns. However, particle improvements have made it possible to pack the SUPELCOGEL H packing material efficiently into conventional 4.6mm ID columns, to improve detection and reduce solvent consumption relative to 7.8mm ID columns.

COLUMN CHARACTERISTICS

Particles:	sulfonated polystyrene/divinylbenzene, spherical, 9 μ m
Counter Ion:	H ⁺
pH Range:	1-13
Organic Compatibility:	<10% in mobile phase
Maximum Flow Rate:	1.5mL/min (7.8mm), 0.4mL/min (4.6mm)
Maximum Pressure:	1000psi (70 bar)

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCOGEL C-610H			
9 μ m	30 x 7.8	59320-U	
SUPELCOGEL H Guard Column (use to protect C-610H)			
9 μ m	5 x 4.6 *	59319	
SUPELCOGEL H			
9 μ m	25 x 4.6	59346	
9 μ m	30 x 7.8	59304-U	
SUPELCOGEL H Guard Column			
9 μ m	5 x 4.6 *	59319	



Organic Acids

Column:	SUPELCOGEL C-610H, 30cm x 7.8mm ID
Cat. No.:	59320
Mobile Phase:	0.1% H_3PO_4
Flow Rate:	0.5mL/min
Temp.:	30°C
Det.:	UV, 210nm
Inj.:	10 μ L

Typical Retention Times for Organic Acids on SUPELCOGEL C-610H and H Columns

COLUMN:	C-610H	H	H
LENGTH:	30cm	30cm	25cm
ID:	7.8mm	7.8mm	4.6mm
CAT. NO.:	59320	59304	59346
Acid	Typical Retention Time (min)		
Acetic	19.0	19.6	17.6
Adipic	22.5	24.0	21.3
Ascorbic	13.1	13.3	12.1
Benzoic ¹	42.4	44.3	37.9
Butyric	28.0	28.3	24.9
Citric	11.0	10.9	10.1
Formic	17.5	18.1	16.3
Fumaric	19.8	20.9	18.2
Gluconic	12.0	12.0	11.1
Isobutyric	25.6	25.9	22.9
Isocitric	11.2	11.0	10.2
Lactic	16.0	16.9	15.2
Maleic	10.4	10.1	9.0
Malic	12.9	13.2	12.0
Malonic	13.4	13.7	12.5
Oxalic	9.0	7.9	7.3
Phytic	8.3	7.0	6.8
Propionic	22.5	23.1	20.5
Quinic	13.3	14.0	12.8
Shikimic	15.5	16.5	14.9
Succinic	15.7	16.4	14.9
Tartaric	11.7	11.7	10.7

Mobile Phase: 0.1% H_3PO_4 , 0.5mL/min (0.17mL/min for 25cm x 4.6mm column), Temperature: 30°C, Detection: UV, 210nm

¹ As sodium benzoate.

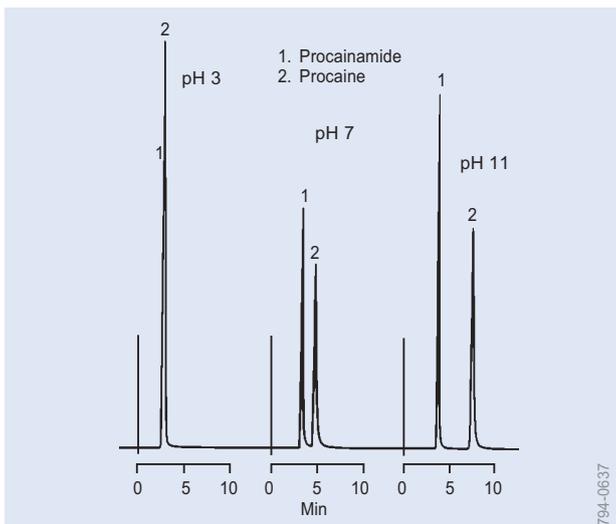
* 5cm x 4.6mm guard column does not include tubing, nuts or ferrules.

HPLC: Small Molecules

Special Purpose SUPELCOGEL Columns

COLUMN CHARACTERISTICS

Packing	
Composition :	polyvinylalcohol polymer, spherical
Functional Groups:	C18
Particle Size:	5µm
Pore Size:	250Å
Surface Area:	150m ² /g
pH:	2-13



794-0637

Separation of Basic Drugs Is Improved at High pH

Column: SUPELCOGEL ODP-50, 15cm x 4.0mm ID, 5µm particles
 Cat. No.: 59307
 Mobile Phase: acetonitrile:25mM phosphate, 40:60
 Flow Rate: 0.45mL/min (850psi/5.8MPa)
 Temp.: 30°C
 Det.: UV, 254nm

Special Purpose Columns: Resin Based

SUPELCOGEL ODP-50: the selectivity of a C18 column, with a wider pH range

Because silica-based bonded phase packings are not stable above pH 7, many basic compounds are analyzed on these columns as positively charged compounds. The charged compounds often interact with residual silanol groups on the packing surface, giving low efficiency and tailing peaks. Resin-based reversed-phase columns are stable over almost the entire pH range, but traditionally have shown low efficiency for most analytes, compared to silica-based columns.

Resin-based SUPELCOGEL ODP-50 columns behave like silica C18 reversed phase columns, but enable you to operate at basic pH (recommended pH range: 2-13). Octadecyl functional groups are covalently attached to the hydroxyl groups of spherical 5µm polyvinylalcohol polymer particles, giving a high density C18 coverage (17% carbon). Separation characteristics are similar to conventional C18 reversed-phase columns – even at high pH. The improved processes used to manufacture SUPELCOGEL ODP-50 particles ensure column efficiencies and mechanical stability that rival those of silica-based packings.

The figure shows the benefits of high pH separations in analysis of basic drugs – at higher pH, the compounds lose their charge and interact more strongly with the packing, prolonging their retention times. Selectivity for most compound pairs is significantly improved, and some pairs reverse their elution order.

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCOGEL ODP-50 (250Å, 17.0% CARBON)			
5µm	15 x 4.0	59307-U	
SUPELCOGEL ODP-50 Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59313C40	
5µm	2 x 4.0 kit**	59312-U	

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

Special Purpose SUPELCOGEL Columns

Special Purpose Columns: Resin Based

SUPELCOGEL TPR-100: a high efficiency resin-based column.

- Unique combination of hydrophilic and hydrophobic (aromatic and aliphatic) monomers
- Crosslinked for mechanical stability
- Narrow pore size and particle size distributions for high efficiency and good peak symmetry
- No micropores
- No bleed from residual porogens
- Use with 100% aqueous or 100% organic mobile phases without swelling or shrinking

Relative to silica, supports composed of organic polymers or resins offer superior pH stability and chemical inertness. Choose a resin-based column when resolution is improved by using a pH above pH 7 or below pH 2, or when silanol or metal ion interactions cause peak tailing or irreproducible retention on a silica-based column.

SUPELCOGEL TPR-100 poly(divinylbenzene/methacrylate) resin has unique selectivity – less hydrophobic than a pure divinylbenzene resin, but less hydrophilic than a methacrylate resin. The resin also offers excellent efficiency, peak symmetry, and mechanical stability. Repeated gradients do not change the bed structure, and the columns can be used with 100% aqueous or 100% organic mobile phases without shrinking or swelling. We recommend using these columns at pressures below 3000 psi.

The figure demonstrates the inertness of SUPELCOGEL TPR-100 resin. Although N,N-dimethylaniline tails severely on the silica-based C18 column, its peak shape on the SUPELCOGEL column is very good, without a competing base modifier in the mobile phase.

PARTICLE SIZE	LENGTH X ID		CAT. NO.	PRICE
	(cm X mm)			
SUPELCOGEL TPR-100 (100Å)				
5µm	15 x 3.0		59154C30	
5µm	15 x 4.0		59154C40	
5µm	15 x 4.6		59154	
SUPELCOGEL TPR-100 Supelguard Cartridges				
5µm	2 x 3.0 (2/pk)		59571C30	
5µm	2 x 4.0 (2/pk)		59571	
5µm	2 x 4.0 kit **		59570-U	

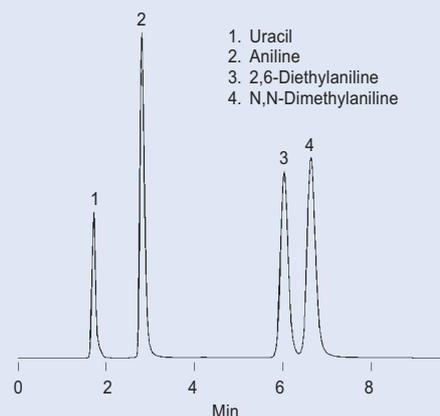
HELPFUL HINTS

Do not use SUPELCOGEL TPR-100 columns with methanol or methanol-containing mixtures.

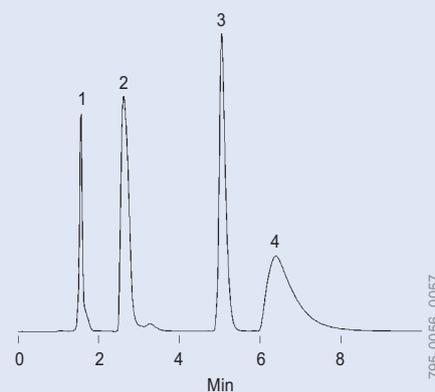
COLUMN CHARACTERISTICS

Particles: poly(divinylbenzene/methacrylate), spherical, 5µm
 Pore Size: 100Å
 Surface Area: 340m²/g
 pH: 2-13

SUPELCOGEL TPR-100 Column



Conventionally Deactivated C18 Column



Better Chromatography of Anilines than on a Deactivated Silica-Based Column

Columns: 15cm x 4.6mm ID, 5µm particles
 Cat. No.: 59154 (SUPELCOGEL TPR-100 Column)
 Mobile Phase: acetonitrile:water, 60:40
 Flow Rate: 1mL/min
 Temp.: ambient
 Det.: UV, 254nm
 Inj.: 10µL

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

Special Purpose SUPELCOSIL & SUPELCOGEL Columns

Special Purpose Columns: Taxols/Taxanes

SUPELCOSIL LC-F columns contain a pentafluorophenyl functional group/encapped packing material. These columns offer selectivities different from traditional reversed phase columns for:

- Halogenated compounds, esters, ketones, and
- Taxanes, including taxol

Taxol, a chemically and pharmacologically unique taxane diterpene amide found in the bark and needles of the Pacific yew tree, has been approved by the US Food and Drug Administration for treatment of ovarian cancer. In isocratic analyses of taxol, the crude and complex sample matrix shortens the lifetimes of traditional reversed phase columns. Analysis of a crude taxol mixture on a SUPELCOSIL LC-F is shown. Excellent resolution of the compounds of interest is obtained by using a mobile phase of acetonitrile, tetrahydrofuran, and water. Small adjustments in the percentage of tetrahydrofuran compensate for normal variation among samples.

COLUMN CHARACTERISTICS

Particles: spherical silica, 5 μ m
 Pore Size: 120Å
 Bonded Phase: pentafluorophenyl
 Surface Area: 170m²/g
 Pore Volume: 0.6mL/g
 pH Range: 2 - 7.5

LENGTH

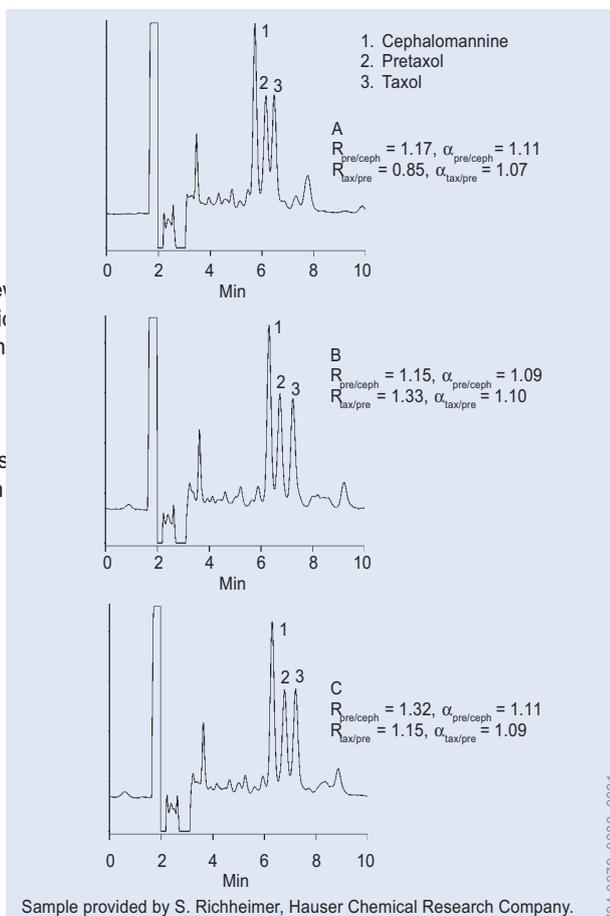
PARTICLE SIZE	(cm X mm)	CAT. NO.	PRICE
SUPELCOSIL LC-F (120Å, 5.0% CARBON)			
5 μ m	25 x 4.0	59158C40	
5 μ m	25 x 4.6	59158	
SUPELCOSIL LC-F Supelguard Cartridges			
5 μ m	2 x 4.0 (2/pk)	59521	
5 μ m	2 x 4.0 kit **	59520	

HELPFUL HINT: PROPERTIES OF ORGANIC SOLVENTS COMMONLY USED IN HPLC

SOLVENT	POLARITY	MISCIBLE WITH WATER?	UV CUTOFF	REFRACTIVE INDEX AT 20°C	SOLVENT STRENGTH, \hat{I}_o (SILICA)	VISCOSITY AT 20°C, CP
Hexane	nonpolar	no	200	1.3750	0.00	0.33
Isooctane		no	200	1.3910	0.01	0.50
Carbon tetrachloride		no	263	1.4595	0.14	0.97
Chloroform		no	245	1.4460	0.31	0.57
Methylene chloride		no	235	1.4240	0.32	0.44
Tetrahydrofuran		yes	215	1.4070	0.35	0.55
Diethyl ether		no	215	1.3530	0.29	0.23
Acetone		yes	330	1.3590	0.43	0.32
Ethyl acetate		poorly	260	1.3720	0.45	0.45
Dioxane		yes	215	1.4220	0.49	1.54
Acetonitrile		yes	190	1.3440	0.50	0.37
2-Propanol		yes	210	1.3770	0.63	2.30
Methanol		yes	205	1.3290	0.73	0.60
Water	polar	yes	-	1.3328	>0.73	1.00

¹ Typical values.

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.



Tetrahydrofuran Concentration Affects Taxol Analysis

Column: SUPELCOSIL LC-F, 25cm x 4.6mm ID, 5 μ m particles
 Cat. No.: 59158
 Mobile Phase: acetonitrile:tetrahydrofuran:water,
 A: 15:30:55, B: 20:25:55, C: 17:28:55
 Flow Rate: 1.5mL/min
 Det.: UV, 227nm
 Inj.: 10 μ L crude taxol mix

HPLC: Small Molecules

SUPELCO SIL Columns

SUPELCO SIL HPLC Columns

Our SUPELCO SIL silica-based HPLC column line includes nearly 30 bonded phase chemistries in a range of particle sizes and column configurations from microbore to preparative scale. When developing a new method, see Supelco's new Discovery suite of reversed phase HPLC columns in this catalog.

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE	PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCO SIL ABZ+PLUS (120Å, 12.0% CARBON)				SUPELCO SIL SUPLEX pKB-100 (120Å, 12.5% CARBON)			
3µm	3.3 x 2.1	5919121		5µm	25 x 2.1	57937	
3µm	5 x 2.1	5919221		5µm	25 x 3.0	58934C30	
3µm	10 x 2.1	57917		5µm	25 x 4.0	58934C40	
3µm	3.3 x 3.0	59191C30		5µm	5 x 4.6	58921-U	
3µm	7.5 x 3.0	59193C30		5µm	15 x 4.6	58932	
3µm	15 x 3.0	59194C30		5µm	25 x 4.6	58934	
3µm	5 x 4.0	59192C40		5µm	25 x 10.0	59172	
3µm	3.3 x 4.6	59191		SUPELCO SIL Suplex pKB-100 Supelguard Cartridges			
3µm	5 x 4.6	59192-U		5µm	2 x 2.1 (2/pk)	59609	
3µm	7.5 x 4.6	59193		5µm	2 x 2.1 kit **	59608	
3µm	15 x 4.6	59194		5µm	2 x 4.0 (2/pk)	59541-U	
5µm	30 x 1.0	57978		5µm	2 x 4.0 kit **	59531-U	
5µm	5 x 2.1	5919521		SUPELCO SIL LC-18 (120Å, 11.0% CARBON)			
5µm	10 x 2.1	57925		3µm	25 x 2.1	57942	
5µm	15 x 2.1	57926		3µm	3.3 x 3.0	58977C30	
5µm	25 x 2.1	57927		3µm	5 x 3.0	58973C30	
5µm	5 x 3.0	59195C30		3µm	15 x 3.0	58985C30	
5µm	15 x 3.0	59196C30		3µm	7.5 x 4.0	58984C40	
5µm	25 x 3.0	59197C30		3µm	15 x 4.0	58985C40	
5µm	5 x 4.0	59195C40		3µm	3.3 x 4.6	58977	
5µm	15 x 4.0	59196C40		3µm	5 x 4.6	58973	
5µm	25 x 4.0	59197C40		3µm	7.5 x 4.6	58984	
5µm	5 x 4.6	59195-U		3µm	15 x 4.6	58985	
5µm	15 x 4.6	59196		5µm	30 x 1.0	57982	
5µm	25 x 4.6	59197		5µm	15 x 2.1	57934	
5µm	25 x 10.0	59179		5µm	25 x 2.1	57935	
5µm	10 x 21.2	59148		5µm	10 x 3.0	59209C30	
5µm	25 x 21.2	54855		5µm	15 x 3.0	58230C30	
12µm	25 x 4.6	59156		5µm	25 x 3.0	58298C30	
12µm	25 x 21.2	59174		5µm	5 x 4.0	58239C40	
SUPELCO SIL ABZ+Plus Supelguard Cartridges				5µm	10 x 4.0	59209C40	
5µm	2 x 2.1 (2/pk)	59605		5µm	15 x 4.0	58230C40	
5µm	2 x 2.1 kit **	59604		5µm	25 x 4.0	58298C40	
5µm	2 x 3.0 (2/pk)	59535C30		5µm	30 x 4.0	59165	
5µm	2 x 4.0 (2/pk)	59535-U		5µm	5 x 4.6	58239	
5µm	2 x 4.0 kit **	59534-U		5µm	10 x 4.6	59209	
SUPELCO SIL LC-ABZ (120Å, 12.0% CARBON)				5µm	15 x 4.6	58230-U	
5µm	30 x 1.0	57990-U		5µm	25 x 4.6	58298	
5µm	25 x 2.1	57936		5µm	25 x 10.0	58368	
5µm	5 x 3.0	59141C30		5µm	25 x 21.2	54849	
5µm	15 x 3.0	59140C30		12µm	25 x 4.6	59182	
5µm	25 x 3.0	59142C30		12µm	25 x 21.2	59185	
5µm	25 x 4.0	59142C40		SUPELCO SIL LC-18 Supelguard Cartridges			
5µm	5 x 4.6	59141		5µm	2 x 2.1 (2/pk)	59613	
5µm	15 x 4.6	59140-U		5µm	2 x 2.1 kit **	59612	
5µm	25 x 4.6	59142		5µm	2 x 3.0 (2/pk)	59564C30	
5µm	25 x 10.0	59170		5µm	2 x 4.0 (2/pk)	59564	
SUPELCO SIL LC-ABZ Supelguard Cartridges				5µm	2 x 4.0 kit **	59554	
5µm	2 x 2.1 (2/pk)	59611					
5µm	2 x 2.1 kit **	59610					
5µm	2 x 3.0 (2/pk)	59545C30					
5µm	2 x 4.0 (2/pk)	59545-U					
5µm	2 x 4.0 kit **	59544-U					

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Small Molecules

SUPELCO SIL Columns

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCO SIL LC-18-DB (120Å, 11.0% CARBON)			
3µm	25 x 2.1	57943	
3µm	3.3 x 3.0	58978C30	
3µm	7.5 x 3.0	58992C30	
3µm	15 x 3.0	58993C30	
3µm	15 x 4.0	58993C40	
3µm	3.3 x 4.6	58978	
3µm	7.5 x 4.6	58992	
3µm	15 x 4.6	58993	
5µm	30 x 1.0	57984	
5µm	25 x 2.1	57940	
5µm	5 x 3.0	58345C30	
5µm	10 x 3.0	59208C30	
5µm	15 x 3.0	58348C30	
5µm	25 x 3.0	58355C30	
5µm	15 x 4.0	58348C40	
5µm	25 x 4.0	58355C40	
5µm	30 x 4.0	59164	
5µm	5 x 4.6	58345	
5µm	10 x 4.6	59208	
5µm	15 x 4.6	58348	
5µm	25 x 4.6	58355-U	
5µm	25 x 10.0	58358	
SUPELCO SIL LC-18-DB Supelguard Cartridges			
5µm	2 x 2.1 (2/pk)	59617	
5µm	2 x 2.1 kit **	59616	
5µm	2 x 3.0 (2/pk)	59565C30	
5µm	2 x 4.0 (2/pk)	59565	
5µm	2 x 4.0 kit **	59555	
SUPELCO SIL LC-18-S (120Å, 11.0% CARBON)			
5µm	30 x 1.0	57920	
5µm	25 x 2.1	57939	
5µm	15 x 3.0	58931C30	
5µm	15 x 4.6	58931	
5µm	25 x 4.6	58928-U	
SUPELCO SIL LC-18-S Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59630	
5µm	2 x 4.0 kit **	59629	
SUPELCO SIL LC-18-T (120Å, 12.3% CARBON)			
3µm	15 x 3.0	58970C30	
3µm	15 x 4.6	58970-U	
5µm	25 x 3.0	58971C30	
5µm	25 x 4.6	58971	
SUPELCO SIL LC-18-T Supelguard Cartridges			
5µm	2 x 3.0 (2/pk)	59621C30	
5µm	2 x 4.0 (2/pk)	59621	
5µm	2 x 4.0 kit **	59620	
SUPELCO SIL LC-DABS (120Å, 12.3% CARBON)			
3µm	15 x 4.6	59137	
SUPELCO SIL LC-18-T Supelguard Cartridges (use for LC-DABS)			
5µm	2 x 4.0 (2/pk)	59621	
5µm	2 x 4.0 kit **	59620	

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCO SIL LC-PAH (120Å)			
3µm	5 x 3.0	59133C30	
3µm	10 x 3.0	59134C30	
3µm	5 x 4.6	59133	
3µm	10 x 4.6	59134	
5µm	25 x 2.1	57945	
5µm	15 x 3.0	58318C30	
5µm	15 x 4.6	58318	
5µm	25 x 4.6	58229	
SUPELCO SIL LC-18 Supelguard Cartridges (use for LC-PAH)			
5µm	2 x 2.1 (2/pk)	59613	
5µm	2 x 2.1 kit **	59612	
5µm	2 x 3.0 (2/pk)	59564C30	
5µm	2 x 4.0 (2/pk)	59564	
5µm	2 x 4.0 kit **	59554	
SUPELCO SIL LC-318 (300Å, 6.0% CARBON)			
5µm	5 x 4.6	58852	
5µm	25 x 4.6	58858	
SUPELCO SIL LC-318 Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59512	
5µm	2 x 4.0 kit **	59502	
SUPELCO SIL LC-8 (120Å, 6.0% CARBON)			
3µm	3.3 x 3.0	58975C30	
3µm	7.5 x 3.0	58982C30	
3µm	15 x 3.0	58983C30	
3µm	15 x 4.0	58983C40	
3µm	3.3 x 4.6	58975	
3µm	7.5 x 4.6	58982	
3µm	15 x 4.6	58983	
5µm	30 x 1.0	57986	
5µm	25 x 2.1	57929	
5µm	15 x 3.0	58220C30	
5µm	25 x 3.0	58297C30	
5µm	15 x 4.0	58220C40	
5µm	25 x 4.0	58297C40	
5µm	5 x 4.6	58238	
5µm	15 x 4.6	58220-U	
5µm	25 x 4.6	58297	
5µm	25 x 10.0	58367	
5µm	25 x 21.2	54845	
12µm	25 x 4.6	59181	
12µm	25 x 21.2	59184	
SUPELCO SIL LC-8 Supelguard Cartridges			
5µm	2 x 2.1 (2/pk)	59615	
5µm	2 x 2.1 kit **	59614	
5µm	2 x 3.0 (2/pk)	59562C30	
5µm	2 x 4.0 (2/pk)	59562	
5µm	2 x 4.0 kit **	59552	

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

HPLC: Small Molecules

SUPELCO SIL Columns

SUPELCO SIL HPLC Columns(cont'd)

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE	PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCO SIL LC-8-DB (120Å, 6.0% CARBON)				SUPELCO SIL LC-CN (120Å, 4.0% CARBON)			
3µm	7.5 x 3.0	58990C30		3µm	3.3 x 3.0	58979C30	
3µm	15 x 3.0	58991C30		3µm	7.5 x 3.0	58986C30	
3µm	15 x 4.0	58991C40		3µm	3.3 x 4.6	58979	
3µm	3.3 x 4.6	58976		3µm	7.5 x 4.6	58986	
3µm	7.5 x 4.6	58990-U		5µm	5 x 3.0	58211C30	
3µm	15 x 4.6	58991		5µm	25 x 3.0	58231C30	
5µm	25 x 2.1	57933		5µm	15 x 4.0	58221C40	
5µm	15 x 3.0	58347C30		5µm	25 x 4.0	58231C40	
5µm	15 x 4.0	58347C40		5µm	5 x 4.6	58211	
5µm	25 x 4.0	58354C40		5µm	15 x 4.6	58221-U	
5µm	5 x 4.6	58344		5µm	25 x 4.6	58231	
5µm	15 x 4.6	58347		5µm	25 x 10.0	58369	
5µm	25 x 4.6	58354		SUPELCO SIL LC-CN Supelguard Cartridges			
5µm	25 x 10.0	58357		5µm	2 x 3.0 (2/pk)	59567C30	
SUPELCO SIL LC-8-DB Supelguard Cartridges				5µm	2 x 4.0 (2/pk)	59567	
5µm	2 x 2.1 (2/pk)	59619		5µm	2 x 4.0 kit **	59557	
5µm	2 x 2.1 kit **	59618		SUPELCO SIL LC-PCN (120Å, 4.0% CARBON)			
5µm	2 x 4.0 (2/pk)	59563		5µm	15 x 3.0	58377C30	
5µm	2 x 4.0 kit **	59553		5µm	25 x 3.0	58378C30	
SUPELCO SIL LC-308 (300Å, 3.5% CARBON)				5µm	15 x 4.0	58377C40	
5µm	5 x 4.6	58851		5µm	15 x 4.6	58377	
5µm	25 x 4.6	58857		5µm	20 x 4.6	59189	
SUPELCO SIL LC-308 Supelguard Cartridges				5µm	25 x 4.6	58378	
5µm	2 x 4.0 (2/pk)	59511-U		SUPELCO SIL LC-PCN Supelguard Cartridges			
5µm	2 x 4.0 kit **	59501		5µm	2 x 4.0 (2/pk)	59514	
SUPELCO SIL LC-DP (120Å, 6.0% CARBON)				5µm	2 x 4.0 kit **	59504	
5µm	15 x 3.0	59150C30		SUPELCO SIL LC-1 (120Å, 2.0% CARBON)			
5µm	25 x 3.0	58842C30		5µm	15 x 3.0	58210C30	
5µm	30 x 4.0	59167		5µm	15 x 4.0	58210C40	
5µm	5 x 4.6	58841		5µm	5 x 4.6	58237	
5µm	10 x 4.6	59211		5µm	15 x 4.6	58210-U	
5µm	15 x 4.6	59150-U		5µm	25 x 4.6	58296	
5µm	25 x 4.6	58842		SUPELCO SIL LC-1 Supelguard Cartridges			
SUPELCO SIL LC-DP Supelguard Cartridges				5µm	2 x 3.0 (2/pk)	59561C30	
5µm	2 x 3.0 (2/pk)	59566C30		5µm	2 x 4.0 (2/pk)	59561	
5µm	2 x 4.0 (2/pk)	59566		SUPELCO SIL LC-NH₂ (120Å, 3.0% CARBON)			
5µm	2 x 4.0 kit **	59556		3µm	7.5 x 3.0	58988C30	
SUPELCO SIL LC-3DP (300Å, 4.0% CARBON)				3µm	15 x 3.0	58989C30	
5µm	25 x 4.6	58859		3µm	7.5 x 4.6	58988	
SUPELCO SIL LC-3DP Supelguard Cartridges				3µm	15 x 4.6	58989	
5µm	2 x 4.0 (2/pk)	59513		5µm	25 x 3.0	58338C30	
SUPELCO SIL LC-F (120Å, 5.0% CARBON)				5µm	25 x 4.0	58338C40	
5µm	25 x 4.0	59158C40		5µm	25 x 4.6	58338	
5µm	25 x 4.6	59158		SUPELCO SIL LC-NH₂ Supelguard Cartridges			
SUPELCO SIL LC-F Supelguard Cartridges				5µm	2 x 3.0 (2/pk)	59568C30	
5µm	2 x 4.0 (2/pk)	59521		5µm	2 x 4.0 (2/pk)	59568	
5µm	2 x 4.0 kit **	59520		5µm	2 x 4.0 kit **	59558	
SUPELCO SIL LC-304 (300Å, 2.7% CARBON)				SUPELCO SIL LC-NH₂-NP (120Å, 3.0% CARBON)			
5µm	5 x 4.6	58823		5µm	25 x 4.6	59132	
5µm	25 x 4.6	58824		SUPELCO SIL LC-NH₂-NP Supelguard Cartridges			
SUPELCO SIL LC-304 Supelguard Cartridges				5µm	2 x 4.0 (2/pk)	59516	
5µm	2 x 4.0 (2/pk)	59592		5µm	2 x 4.0 kit **	59515	
5µm	2 x 4.0 kit **	59591					

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Small Molecules

SUPELCO SIL & SUPELCO GEL Columns

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCO SIL HISEP (120Å)			
5µm	25 x 2.1	57932	
5µm	5 x 4.6	59143	
5µm	15 x 4.6	58935	
5µm	25 x 4.6	58919	
SUPELCO SIL Hisep Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59640-U	
5µm	2 x 4.0 kit **	59639	
SUPELCO SIL LC-Si (120Å)			
3µm	7.5 x 3.0	58980C30	
3µm	15 x 3.0	58981C30	
3µm	15 x 4.0	58981C40	
3µm	3.3 x 4.6	58974	
3µm	7.5 x 4.6	58980-U	
3µm	15 x 4.6	58981	
5µm	30 x 1.0	57980-U	
5µm	25 x 2.1	57930-U	
5µm	10 x 3.0	59210C30	
5µm	15 x 3.0	58200C30	
5µm	15 x 4.0	58200C40	
5µm	25 x 4.0	58295C40	
5µm	30 x 4.0	59166	
5µm	5 x 4.6	58236	
5µm	10 x 4.6	59210-U	
5µm	15 x 4.6	58200-U	
5µm	25 x 4.6	58295	
5µm	25 x 10.0	58365	
5µm	25 x 21.2	54843	
12µm	25 x 4.6	59180-U	
12µm	25 x 21.2	59183	
SUPELCO SIL LC-Si Supelguard Cartridges			
5µm	2 x 3.0 (2/pk)	59560C30	
5µm	2 x 4.0 (2/pk)	59560	
5µm	2 x 4.0 kit **	59550	
SUPELCO SIL LC-3Si (300Å)			
5µm	25 x 6.2	58965	
SUPELCO SIL LC-Si Supelguard Cartridges (use for LC-3Si)			
5µm	2 x 4.0 (2/pk)	59560	
5µm	2 x 4.0 kit **	59550	
SUPELCO SIL LC-DIOL (120Å, 3.5% CARBON)			
5µm	25 x 3.0	58201C30	
5µm	25 x 4.0	58201C40	
5µm	25 x 4.6	58201	
SUPELCO SIL LC-Diol Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59569	
5µm	2 x 4.0 kit **	59559	
SUPELCO SIL SAX1 (120Å)			
5µm	25 x 3.0	59138C30	
5µm	25 x 4.0	59138C40	
5µm	25 x 4.6	59138	
SUPELCO SIL SAX1 Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59537-U	
5µm	2 x 4.0 kit **	59536-U	
SUPELCO SIL LC-SCX (120Å)			
5µm	25 x 3.0	58997C30	
5µm	25 x 4.6	58997	
SUPELCO SIL LC-SCX Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59519	
5µm	2 x 4.0 kit **	59509	

** Kits include one cartridge, stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

**** Does not include tubing, nuts or ferrules.

SUPELCO GEL resin-based HPLC Columns

For reversed-phase separations at high pH or low pH, we offer SUPELCO GEL TPR-100 and SUPELCO GEL ODP-50 resin-based HPLC columns. SUPELCO GEL resin-based ion exclusion HPLC columns contain sulfonated divinylbenzene resins in six cationic forms, each offering a unique selectivity for analyses of saccharides or organic acids.

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
SUPELCO GEL TPR-100 (100Å)			
5µm	15 x 3.0	59154C30	
5µm	15 x 4.0	59154C40	
5µm	15 x 4.6	59154	
SUPELCO GEL TPR-100 Supelguard Cartridges			
5µm	2 x 3.0 (2/pk)	59571C30	
5µm	2 x 4.0 (2/pk)	59571	
5µm	2 x 4.0 kit **	59570-U	
SUPELCO GEL ODP-50 (250Å, 17.0% CARBON)			
5µm	15 x 4.0	59307-U	
SUPELCO GEL ODP-50 Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59313C40	
5µm	2 x 4.0 kit **	59312-U	
SUPELCO GEL AG1			
9µm	30 x 7.8	59318-U	
SUPELCO GEL AG1 Guard Column			
9µm	5 x 4.6 ****	59317-U	
SUPELCO GEL AG2			
9µm	30 x 7.8	59315	
SUPELCO GEL AG2 Guard Column			
9µm	5 x 4.6 ****	59316	
SUPELCO GEL C-610H			
9µm	30 x 7.8	59320-U	
SUPELCO GEL H Guard Column (use to protect C-610H)			
9µm	5 x 4.6 ****	59319	
SUPELCO GEL C-611			
9µm	30 x 7.8	59310-U	
SUPELCO GEL Ca Guard Column (use to protect C-611)			
9µm	5 x 4.6 ****	59306-U	
SUPELCO GEL CA			
9µm	30 x 7.8	59305-U	
SUPELCO GEL Ca Guard Column			
9µm	5 x 4.6 ****	59306-U	
SUPELCO GEL H			
9µm	25 x 4.6	59346	
9µm	30 x 7.8	59304-U	
SUPELCO GEL H Guard Column			
9µm	5 x 4.6 ****	59319	
SUPELCO GEL K			
9µm	30 x 7.8	59342	
SUPELCO GEL K Guard Column			
9µm	5 x 4.6 ****	59344	
SUPELCO GEL Pb			
9µm	30 x 7.8	59343	
SUPELCO GEL Pb Guard Column			
9µm	5 x 4.6 ****	59345	

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

Liquid
Chromatography

SUPELCO

HPLC: Small Molecules

Other Columns

Other HPLC Columns - Small Molecules

In addition to our own product lines, we offer name brand silica-based columns: alphaBond, Hypersil, Kromasil, LiChrosorb, LiChrospher, Nucleosil, TSK-GEL, and Waters Spherisorb.

alphaBond

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
ALPHABOND (125Å)			
C18			
10µm	15 x 3.9	57488	
10µm	30 x 3.9	57489	
alphaBond Guard Cartridges			
C18			
10µm	1 x 4.6 (4/pk)	57490-U	

Hypersil

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
HYPERASIL (120Å)			
ODS (C18)			
3µm	10 x 3.2	54931	
3µm	10 x 4.6	Z226327	
5µm	15 x 3.2	54932	
5µm	25 x 3.2	54933	
5µm	15 x 4.6	Z226335	
5µm	25 x 4.6	Z226343	
BDS-C18*			
5µm	15 x 3.2	54915-U	
5µm	25 x 3.2	54916	
5µm	15 x 4.6	57485	
5µm	25 x 4.6	57486	
MOS (C8)			
3µm	10 x 3.2	54934	
3µm	10 x 4.6	Z226351	
5µm	15 x 4.6	Z226378	
5µm	25 x 4.6	Z226386	
BDS-C8*			
5µm	15 x 4.6	506109	
5µm	25 x 4.6	506095	
Phenyl			
3µm	10 x 3.2	54943	
3µm	10 x 4.6	Z226459	
5µm	25 x 3.2	54945	
5µm	15 x 4.6	Z226467	
5µm	25 x 4.6	Z226475	
CPS (Cyano)			
3µm	10 x 3.2	54940	
3µm	10 x 4.6	Z226424	
5µm	25 x 3.2	54942	
5µm	15 x 4.6	Z226432	
5µm	25 x 4.6	Z226440	
SAS (C1)			
3µm	10 x 4.6	Z226483	
5µm	15 x 3.2	54947-U	
5µm	15 x 4.6	Z226491	
5µm	25 x 4.6	Z226505	

* BDS = Base Deactivated Silica.

** Deactivated C18.

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
HYPERASIL (120Å) CONT'D			
Silica			
3µm	10 x 4.6	Z226297	
5µm	15 x 4.6	Z226300	
5µm	25 x 4.6	Z226319	
APS (NH ₂)			
3µm	10 x 3.2	54937	
3µm	10 x 4.6	Z226394	
5µm	15 x 3.2	54938	
5µm	25 x 3.2	54939	
5µm	15 x 4.6	Z226408	
5µm	25 x 4.6	Z226416	
Hypersil Guard Cartridges			
ODS (C18)			
5µm	1 x 4.6 (4/pk)	Z227064	
BDS-C18*			
5µm	1 x 4.6 (4/pk)	57487	
MOS (C8)			
5µm	1 x 4.6 (4/pk)	Z227072	
BDS-C8*			
5µm	1 x 4.6 (4/pk)	506117	
Phenyl			
5µm	1 x 4.6 (4/pk)	Z227102	
CPS (Cyano)			
5µm	1 x 4.6 (4/pk)	Z227099	
SAS (C1)			
5µm	1 x 4.6 (4/pk)	Z227110	
Silica			
5µm	1 x 4.6 (4/pk)	Z227056	
APS (NH ₂)			
5µm	1 x 4.6 (4/pk)	Z227080	

Inertsil

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
INERTSIL (150Å)			
ODS-2**			
5µm	15 x 4.6	506079	
5µm	25 x 4.6	506060	
Inertsil Guard Cartridges			
ODS-2**			
5µm	1 x 4.6 (4/pk)	506087	

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Small Molecules Other Columns

49

Liquid
Chromatography

Kromasil

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
KROMASIL (100Å)			
C18			
5µm	15 x 4.6	55148	
5µm	25 x 4.6	55147	
10µm	25 x 4.6	55149	
C8			
5µm	15 x 4.6	55151	
5µm	25 x 4.6	55150-U	
10µm	25 x 4.6	55152	
C4			
5µm	15 x 4.6	55154-U	
5µm	25 x 4.6	55153	
60-A (Silica, 60Å)			
5µm	15 x 4.6	55163	
5µm	25 x 4.6	55162	
10µm	25 x 4.6	55164	
100-A (Silica)			
5µm	25 x 4.6	55159	
10µm	25 x 4.6	55161	
NH₂			
5µm	25 x 4.6	55156-U	
10µm	25 x 4.6	55158-U	
Kromasil Guard Cartridges			
C18			
5µm	1 x 4.6 (4/pk)	55165-U	
C8			
5µm	1 x 4.6 (4/pk)	55166	
C4			
5µm	1 x 4.6 (4/pk)	55167	
60-A (Silica, 60Å)			
5µm	1 x 4.6 (4/pk)	55170-U	

Lichrosorb

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
LICHROSORB (100Å)			
RP-18 (C18)			
5µm	15 x 3.2	54952	
5µm	25 x 3.2	54950-U	
5µm	15 x 4.6	54951	
5µm	25 x 4.6	54949	
RP-8 (C8)			
5µm	15 x 4.6	54955-U	
5µm	25 x 4.6	54953-U	
Si-60 (Silica, 60Å)			
5µm	25 x 3.2	54962	
5µm	15 x 4.6	54963-U	
5µm	25 x 4.6	54961	
NH₂			
5µm	25 x 3.2	54958-U	
5µm	15 x 4.6	54959	
5µm	25 x 4.6	54957-U	

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
LiChrosorb Guard Cartridges			
RP-18 (C18)			
5µm	1 x 4.6 (4/pk)	54965-U	
RP-8 (C8)			
5µm	1 x 4.6 (4/pk)	54966	
Si-60 (Silica, 60Å)			
5µm	1 x 4.6 (4/pk)	54968	
NH₂			
5µm	1 x 4.6 (4/pk)	54967	

Licrospher

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
LICHROSPHER (100Å)			
RP-18 (C18)			
5µm	15 x 3.2	54775	
5µm	25 x 3.2	54777	
5µm	15 x 4.6	54774	
5µm	25 x 4.6	54776	
RP-8 (C8)			
5µm	15 x 3.2	54779	
5µm	15 x 4.6	54778	
5µm	25 x 4.6	54780	
CN (Cyano)			
5µm	15 x 4.6	54786	
5µm	25 x 4.6	54788	
Si-60 (Silica, 60Å)			
5µm	15 x 3.2	54791-U	
5µm	15 x 4.6	54790-U	
5µm	25 x 4.6	54792	
NH₂			
5µm	15 x 3.2	54783	
5µm	25 x 3.2	54785	
5µm	15 x 4.6	54782	
5µm	25 x 4.6	54784	
LiChrospher Guard Cartridges			
RP-18 (C18)			
5µm	1 x 4.6 (4/pk)	54794	
CN (Cyano)			
5µm	1 x 4.6 (4/pk)	54798	
Si-60 (Silica, 60Å)			
5µm	1 x 4.6 (4/pk)	54797-U	
NH₂			
5µm	1 x 4.6 (4/pk)	54796-U	

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

SUPELCO

HPLC: Small Molecules

Other Columns

Nucleosil

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
NUCLEOSIL (100Å)			
C18			
3µm	10 x 3.2	54917	
3µm	10 x 4.6	Z226165	
5µm	15 x 3.2	54918	
5µm	25 x 3.2	54919	
5µm	15 x 4.6	Z226173	
5µm	25 x 4.6	Z226181	
C8			
3µm	10 x 3.2	54920-U	
3µm	10 x 4.6	Z226203	
5µm	15 x 3.2	54921	
5µm	25 x 3.2	54922	
5µm	15 x 4.6	Z226211	
5µm	25 x 4.6	Z226238	
Phenyl			
7µm	25 x 4.6	Z226246	
CN (Cyano)			
5µm	15 x 3.2	54924	
5µm	25 x 3.2	54925-U	
5µm	15 x 4.6	Z226254	
5µm	25 x 4.6	Z226262	
Silica			
5µm	25 x 3.2	54914	
5µm	15 x 4.6	Z226149	
5µm	25 x 4.6	Z226157	
NH ₂			
5µm	15 x 3.2	54926	
5µm	25 x 3.2	54927	
5µm	25 x 4.6	Z226289	
Nucleosil Guard Cartridges			
C18			
5µm	1 x 4.6 (4/pk)	Z227137	
C8			
5µm	1 x 4.6 (4/pk)	Z227145	
Phenyl			
7µm	1 x 4.6 (4/pk)	Z227153	
CN (Cyano)			
5µm	1 x 4.6 (4/pk)	Z227161	
Silica			
5µm	1 x 4.6 (4/pk)	Z227129	
NH ₂			
5µm	1 x 4.6 (4/pk)	Z227188	

TSK-GEL C18

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
TSK-GEL C18 COLUMNS			
ODS-80Tm (80Å)			
5µm	7.5 x 4.6	816651	
5µm	15 x 4.6	808148	
5µm	25 x 4.6	808149	
ODS-80Ts (80Å)			
5µm	7.5 x 4.6	817200	
5µm	15 x 4.6	817201	
5µm	25 x 4.6	817202	
ODS-120A (120Å)			
5µm	15 x 4.6	807636	
5µm	25 x 4.6	807124	
ODS-120T (120Å)			
5µm	15 x 4.6	807637	
5µm	25 x 4.6	807125	

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
TSK-GEL C18 COLUMNS, CONT'D			
Super-ODS (110Å)			
2µm	5 x 4.6	818154	
2µm	10 x 4.6	818197	
TSK-GEL Guard Cartridges and Filters			
ODS-80Ts (80Å)			
5µm	1.5 x 3.2 (3/pk)	817242	
ODS-120T (120Å)			
5µm	1.5 x 3.2 (3/pk)	814125	
Filter			
	(3/pk)	818207	

Waters Spherisorb

PARTICLE SIZE	LENGTH X ID (cm X mm)	CAT. NO.	PRICE
WATERS SPHERISORB (80Å)			
ODS-2 (C18)			
3µm	10 x 3.2	54903	
3µm	10 x 4.6	Z226033	
5µm	15 x 3.2	54904	
5µm	25 x 3.2	54905	
5µm	15 x 4.6	Z226041	
5µm	25 x 4.6	Z226068	
Octyl (C8)			
3µm	10 x 4.6	Z226076	
5µm	25 x 3.2	54908	
5µm	15 x 4.6	Z226084	
5µm	25 x 4.6	Z226092	
Phenyl			
5µm	25 x 4.6	Z226106	
Cyano			
5µm	25 x 3.2	54910	
5µm	25 x 4.6	Z226114	
Silica			
3µm	10 x 4.6	Z226009	
5µm	15 x 3.2	54901	
5µm	25 x 3.2	54902	
5µm	15 x 4.6	Z226017	
5µm	25 x 4.6	Z226025	
Amino (NH ₂)			
5µm	25 x 3.2	54911-U	
5µm	25 x 4.6	Z226122	
SAX			
5µm	25 x 4.6	Z226130	
Waters Spherisorb Guard Cartridges			
ODS-2 (C18)			
5µm	1 x 4.6 (4/pk)	Z226971	
Octyl (C8)			
5µm	1 x 4.6 (4/pk)	Z226998	
Phenyl			
5µm	1 x 4.6 (4/pk)	Z227005	
Cyano			
5µm	1 x 4.6 (4/pk)	Z227013	
Silica			
5µm	1 x 4.6 (4/pk)	Z226963	
Amino (NH ₂)			
5µm	1 x 4.6 (4/pk)	Z227021	
SAX			
5µm	1 x 4.6 (4/pk)	Z227048	

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Small Molecules Guard Column Holders



Guard Column Holders

P000834

Use these guard column holders with the guard cartridges listed on the previous pages. The Direct-Connect holders allow a guard cartridge to attach to a Supelco modular column with no dead volume. The Direct-Connect holders can only be used with Supelco modular columns. The Swivel-type holders allow the tubing to move independently of the holder, reducing the risk of leaks caused by crimped tubing. The Stand-Alone holders include the necessary tubing, nuts and ferrules for connecting to any analytical columns.

DESCRIPTION	CAT. NO.	PRICE
FOR SUPELCO SUPELGUARD CARTRIDGES		
Direct-Connect (Swivel-type) Holder*	504262	
Direct-Connect (Swivel-type) Holder**	504254	
Direct-Connect Holder**	55205	
Stand-Alone Holder***	59660-U	
Stand-Alone Holder****	567499-U	
FOR SUPELCO PELLIGUARD CARTRIDGES		
Stand-Alone Holder	500054	
FOR OTHER CARTRIDGES		
Stand-Alone Holder (includes PEEK Column Coupler)	54987	
Replacement PEEK Column Coupler	54986	
FOR TSK-GEL CARTRIDGES AND FILTERS		
Stand-Alone Holder for TSK-GEL Cartridges	814100	
Stand-Alone Holder for TSK-GEL Filters	818206	

* For 2.1mm ID Supelco columns

** For 3.0, 4.0 and 4.6mm ID Supelco columns

*** For 2.1, 3.0, 4.0 and 4.6mm ID Supelco columns

**** For 10.0mm ID Supelco columns

HELPFUL HINTS

Before flushing a reversed-phase HPLC column that contains a buffer (salt), flush with warm (60°C) DI H₂O thoroughly to remove salts. Not following this general rule may result in salt precipitation when returned to 100% organic for long-term storage. For more information, refer to literature T401012, Buffer Solubility section.

Pelliguard Guard Cartridges

For 5µm, 10µm, or 12µm SUPELCOSIL and other silica-based HPLC columns, where samples are especially dirty, and a small loss of efficiency is acceptable. Each kit contains one cartridge (2cm x 4.6mm ID) filled with 40µm Pelliguard packing, a reusable stand-alone column holder, and hardware for connecting the holder to 1/16" tubing. Replacement cartridges come in packages of four.

COLUMN TO BE PROTECTED	RECOMMENDED PELLIGUARD COLUMN	CAT. NO.	PRICE
Silica	LC-Si Kit	59641	
	Cartridges (pk. of 4)	59651	
Cyano	LC-CN Kit	59645-U	
	Cartridges (pk. of 4)	59655	
Amino	LC-NH ₂ Kit	59646	
	Cartridges (pk. of 4)	59656	
C8	LC-8 Kit	59643	
	Cartridges (pk. of 4)	59653	
C18	LC-18 Kit	59644	
	Cartridges (pk. of 4)	59654	



Bulk Pellicular Packing Kits

Reusable 5cm x 4.6mm ID guard column hardware and 40µm pellicular packing, for protecting 10µm columns. Each column kit contains an empty 5cm x 4.6mm ID column, 10g of Pelliguard packing, 10 frits, and hardware for connecting the column to 1/16" tubing. About 1.3 grams of packing is needed to pack one 5cm x 4.6mm column.

COLUMN TO BE PROTECTED	RECOMMENDED PELLIGUARD COLUMN	CAT. NO.	PRICE
Silica	LC-Si Kit	58202	
	LC-Si Packing, 10g	58291	
Cyano	LC-CN Kit	58234	
	LC-CN Packing, 10g	58235	
C8	LC-8 Kit	58222-U	
	LC-8 Packing, 10g	58293	
C18	LC-18 Kit	58232	
	LC-18 Packing, 10g	58294	

Guard Column Hardware Kit, Funnel and Tubing

Kit includes 5cm x 4.6mm ID column, endfittings, 2 frits (2.0µm pores), and 2"/5cm of 0.01" ID x 1/16" OD SS tubing. Funnel connects to column with tygon tubing (included) for easier column filling.

DESCRIPTION	CAT. NO.	PRICE
Guard Column Hardware Kit	58319	
Replacement Frits (pk. of 10)	58264	
Funnel and Tubing	20390-U	



HPLC: Small Molecules

HPLC Column Test Mixes

HPLC Column Test Mixes

Performance evaluation mixes for HPLC columns.

Well defined test mixes enable you to troubleshoot chromatographic problems, optimize system efficiency, and evaluate columns under conditions where their performance is understood. We ship these test mixes in amber ampuls to prevent photodegradation, and we include instructions for proper use and interpretation of results.

Choose from column-specific or application-specific mixes. All mixes except the amino phase test mix (Cat. No. 58424) call for detection; the amino phase test mix (sugars) calls for refractive index detection. We recommend our HPLC Troubleshooting Guide (Bulletin 826) for additional information about using test mixes.

1mL unless otherwise specified.

TEST MIX	USE TO TEST	SOLVENT	COMPONENTS (CONC./mL)	CAT. NO.	PRICE
Amino Phase	LC-NH ₂ columns	acetonitrile:water, 25:75	D-fructose (25mg) α-D-glucose (25mg) sucrose (25mg) maltose (25mg) lactose (25mg)	58424	
Cyano Phase	LC-CN, LC-PCN, ABZPlus columns, any weakly hydrophobic phase	acetonitrile:water, 25:75	uracil (7μg) acetophenone (7μg) benzene (750μg) toluene (775μg)	58299	
Normal Phase Mix 1	LC-Si (silica) columns	methylene chloride	benzene (600μg) benzanilide (20μg) acetanilide (20μg)	58281	
Normal Phase Mix 2	LC-Si, LC-CN, LC-NH ₂ columns	ethanol:hexane, 5:95	toluene (1mg) diethyl phthalate (1mg) dimethyl phthalate (1mg)	47640-U	
Nucleosides	LC-18-S columns	water, sodium formate (10mg/mL)	12 nucleosides (10-100μg) (see page 146)	47310-U	
LC-PAH	LC-PAH columns	methanol: methylene chloride, 50:50	16 PAHs (100-2000μg) (see page 410)	48743	
Peptide Standard	reversed phase columns used for peptide separations (e.g., 300Å phases)	none (dried film)	Gly-Tyr (~0.125mg) Val-Tyr-Val (~0.5mg) Met enkephalin (~0.5mg) Leu enkephalin (~0.5mg) angiotensin II (~0.5mg)	H2016-1VL	
Reversed Phase Mix 1	hydrophobic RP columns (e.g., LC-8, LC-18)	methanol:water, 60:40	uracil (7μg) acetophenone (7μg) benzene (750μg) toluene (775μg)	58278	
Reversed Phase Mix 2	hydrophobic RP columns (e.g., LC-8, LC-18)	acetonitrile:water, 58:42	uracil (5μg) phenol (700μg) N,N-diethyl-m-toluamide (600μg) toluene (4mg)	47641-U	
Chiral 1	chiral phases	hexane:ethyl acetate, 80:20	toluene (70μg) (+) TFAE (25μg) (-) TFAE (25μg)	48250-U	
Chiral 2	chiral phases	chloroform	benzene (500μg) (+) N-PDBA ² (50μg) (-) N-PDBA ² (50μg)	48251	

¹ 2,2,2-trifluoro-1-(9-anthryl)ethanol

² N-(1-phenylethyl)-3,5-dinitrobenzamide

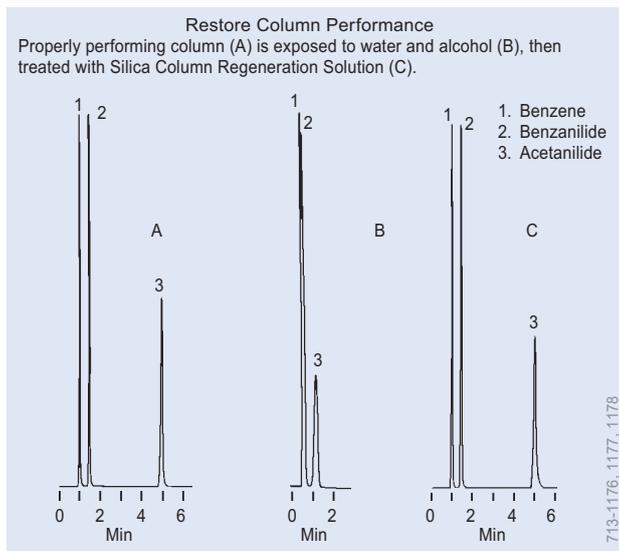
Custom Test Mixes

For information on made-to-order standards and test mixes, call our Technical Service chemists, or request our Custom Chemical Reference brochure (Publication No. 196905).

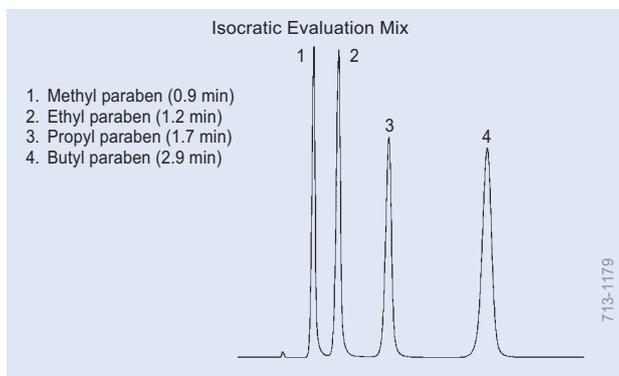
Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Small Molecules

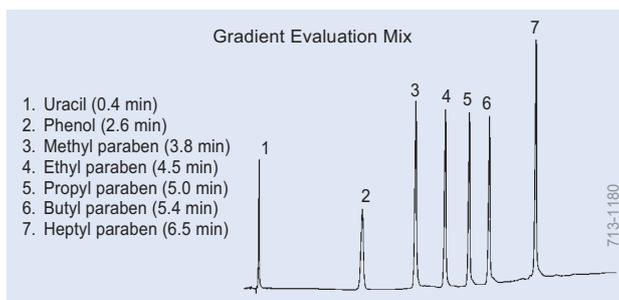
HPLC Column Test Mixes



Column: SUPELCO SIL LC-Si, 15cm x 4.6mm, 3 μ m particles
 Cat. No.: 58981
 Mobile Phase: A = methylene chloride:methanol:water (99.4:0.5:0.1)
 B = 2-propanol:water (50:50)
 C = Silica Column Regeneration Solution, 4mL/min for 10 min,
 then methylene chloride:methanol:water (99.4:0.5:0.1), 2mL/min
 for 10 min
 Det.: UV, 254nm
 Inj.: 10 μ L



Flow Rate: 2mL/min
 Det.: UV, 254nm
 Inj.: 10 μ L



Flow Rate: 2mL/min methanol:water, 10:90 to 90:10 in 5 min
 Det.: UV, 254nm
 Inj.: 10 μ L

HPLC Column Test Mixes

System diagnostics: test kits and column regeneration solutions.

Silica Column Regeneration Solution This solution effectively regenerates a silica column that has come into contact with very strongly polar solvents, such as water or alcohols. Simply flush the column with regeneration solution for 10 minutes, then reequilibrate with mobile phase for 10 minutes. Column performance usually is restored to that obtained before exposure to the polar solvent.

System Diagnostic Kit -Take a systematic approach to diagnosing problems in an HPLC system. This kit consists of:

- 5cm x 4.6mm SUPELCO SIL LC-18SD column
- 6 x 1mL Isocratic Evaluation Mix
- 6 x 1mL Gradient Evaluation Mix
- four 1.8mL screwcap vials with hole caps and septa

When you need to determine the cause of a problem, install the 5cm column, prepare a simple methanol:water mobile phase, and inject 10 μ L of Isocratic Evaluation Mix onto the column. Compare your chromatogram with that from a properly performing system and use the information sheet included with the kit to help isolate the source of the problem. If necessary, make injections with the gradient mix.

We recommend our HPLC Troubleshooting Guide (Bulletin 826, available free on request) to help you interpret the results you obtain.

Evaluation Test Mixes -Six 1mL ampuls of test compounds in methanol:water, 60:40.

These formulations are designed for evaluating how reliably a chromatographic system is providing such fundamentally important parameters as flow rate, proportioning, and mixing.

DESCRIPTION	QTY.	CAT. NO.	PRICE
Silica Column Regeneration Solution	200mL	33175	
LC-18SD System Diagnostic Kit		58543	
Isocratic Evaluation Mix	6 x 1mL	48270-U	
Gradient Evaluation Mix	6 x 1mL	48271	

RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No.	Subject
T100826	HPLC troubleshooting guide
T196905	custom chemicals brochure

HPLC: Biopolymers

Column Selection: Nucleic Acid Separations

SAMPLE TYPE	SEPARATION MODE	MCI GEL HPLC (SEE PAGE)	AMERSHAM BIOSCIENCES FPLC (SEE PAGE)	TSK-GEL HPLC (SEE PAGE)	SUPELCO HPLC (SEE PAGE)
DNA/RNA	Gel Filtration		Superdex 200 (62)	G-DNA-PW (63)	
	Ion Exchange	ProtEx-DEAE (60)	Mono Q (60)	DEAE-NPR (61)	
PCR Fragments	Ion Exchange	ProtEx-DEAE (60)	Mono Q (60)	DEAE-NPR (61)	
Oligonucleotides	Ion Exchange	ProtEx-DEAE (60)	Mono Q (60)	DEAE-5PW (61) DEAE-NPR (61)	
	Reversed Phase			Oligo-DNA RP (64)	SUPELCOSIL LC-318 (64) Discovery C18 (28)
Nucleotides	Reversed-Phase				SUPELCOSIL LC-18-T (39) Discovery C18 (28) Discovery RP-AmideC16 (29)
	Ion Exchange			DEAE-2SW (61)	SUPELCOSIL SAX1 (47)
Nucleosides	Reversed-Phase				SUPELCOSIL LC-18-S (39) Discovery C18 (28) Discovery RP-AmideC16 (29)
Nucleic Acid Bases	Reversed-Phase				SUPELCOSIL LC-18 (44) SUPELCOSIL LC-18-DB (33) Discovery C18 (28) Discovery RP-AmideC16 (29)
	Ion Exchange				SUPELCOSIL LC-SCX (47)

RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No. Subject
T402038 Discovery BIO Wide Pore HPLC Columns and Capillaries: Solutions to Protein and Peptide Separation Challenges

McClung, G., W.T. Frankenburger, Jr. Comparison of Reversed Phase High Performance Liquid Chromatographic Methods for Precolumn Derivatized Amino Acids. *J. Liq. Chromatogr.* 11: 613-646 (1988). Reference not available from Supelco.

HPLC: Biopolymers

Column Selection: Peptide and Protein Separations

SEPARATION MODE	MCI GEL HPLC (SEE PAGE)	AMERSHAM BIOSCIENCES FPLC (SEE PAGE)	TSK-GEL HPLC (SEE PAGE)	SUPELCO HPLC (SEE PAGE)
Gel Filtration		Superdex Peptide Superdex 75 Superdex 200 (62) Fast Desalting HR 10/10 (62)	G2000SW, G2000SW _{XL} G3000SW, G3000SW _{XL} G4000SW, G4000SW _{XL} Super 3000 SW (62)	
Strong Anion Exchange	ProtEx-DEAE (60)	Mono Q (60)		
Weak Anion Exchange		Mono P (60)	DEAE-5PW, DEAE-NPR (61)	
Chromatofocusing		Mono P (60)		
Strong Cation Exchange	ProtEx-SP (60)	Mono S (60)	SP-5PW, SP-NPR (61)	
Weak Cation Exchange			CM-5PW (61)	
Hydrophobic Interaction			Ether-5PW Phenyl-5PW Butyl-NPR (64)	
Reversed Phase			Phenyl-5PW RP Octadecyl-4PW Octadecyl-NPR Super-ODS (64)	Discovery BIO Wide Pore C18 (57) Discovery BIO Wide Pore C8 (58) Discovery BIO Wide Pore C5 (59) Discovery RP-AmideC16 (29) Discovery C18 (28) Discovery C8 (30)
Affinity			ABA-5PW Boronate-5PW Chelate-5PW Heparin-5PW Tresyl-5PW (64)	

Note: Products in italics are used under low pressure conditions.

Columns for Amino Acid Separations

DERIVATIZED AMINO ACID	SUPEL COSIL HPLC	DIMENSIONS (cm x mm ID)		PRICE	SUPEL GUARD GUARD COLUMN	CAT. NO.	PRICE
Dabsyl-AA	LC-DABS (3µm)	15 x 2.1	59137		LC-18-T	59621	
DABTH-AA	LC-18 (3µm)	15 x 4.6	58985		LC-18	59564	
OPA-AA	LC-18 (5µm)	15 x 4.6	58230-U		LC-18	59564	
PTC-AA	LC-18-DB (5µm)	25 x 4.6	58355-U		LC-18-DB	59565	
PTH-AA	LC-18-DB (3µm)	25 x 2.1	57943		LC-18-DB	59565	
	LC-18-DB (5µm) ¹	25 x 4.6	58355-U		LC-18-DB	59565	

¹ Alternative to 3µm LC-18-DB.

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

Liquid Chromatography

SUPELCO

HPLC: Biopolymers

Discovery BIO Wide Pore Columns

Discovery BIO

Solutions to Protein and Peptide Separation Challenges

Discovery BIO HPLC columns and capillaries provide sensitive, stable, efficient, reproducible separations of proteins and peptides. The different phase chemistries provide unique selectivity increasing your resolution options. Separations are completely scalable from analytical to prep. The low-bleed feature and microbore and capillary dimensions make them ideal for proteomics and LC/MS applications.



Discovery BIO Wide Pore C18

Ideal for high resolution peptide mapping and synthetic and native peptide separations.

Discovery BIO Wide Pore C8

Also ideal for high resolution peptide mapping and synthetic and native peptide separations. The intermediate hydrophobicity of the C8 phase makes it very useful for method development.

Discovery BIO Wide Pore C5

The short-chain C5 phase is used for protein and hydrophobic peptide separations. Discovery BIO Wide Pore C5 is more stable than the commonly-used C4 phases.

Capillary and Microbore Dimensions

With the rapid growth of proteomics, the demand for detailed characterization on smaller sample volumes is increasing. So is the utility of smaller ID HPLC columns. Traditional column IDs of 4.6mm and even 2.1mm are being replaced by microbore (1mm ID) and capillary (< 1mm ID) dimensions. The benefits are two-fold: increased sensitivity and decreased sample consumption. Discovery BIO C18 and C5 is available in capillary and microbore dimensions to meet these criteria.

Go to www.sigma-aldrich.com/supelco-bio to see the entire Discovery BIO line, including new additions, and download the most recent literature and applications.

Mixture of Synthetic Peptides on Discovery BIO Wide Pore C18 and a Leading Competitive Column

Columns: (A) Discovery BIO Wide Pore C18, 15cm x 4.6mm, 5µm (Cat. No. 568222-U)

(B) Competitive protein and peptide C18, 15cm x 4.6mm, 300Å, 5µm

Mobile Phase: (A) 80:20, (0.1% TFA in water): (0.1% TFA in CH₃CN); (B) 66:34, (0.1% TFA in water): (0.1% TFA in CH₃CN)

Flow Rate: 1.0mL/min

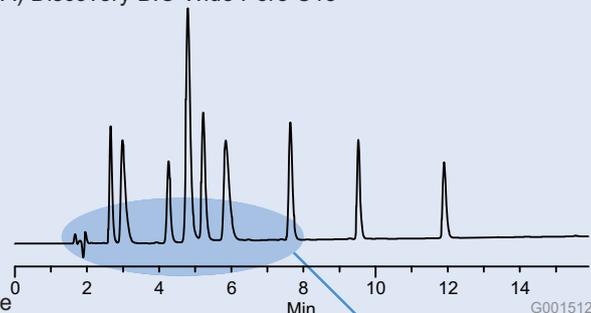
Temp: 30°C

Detection: 220nm

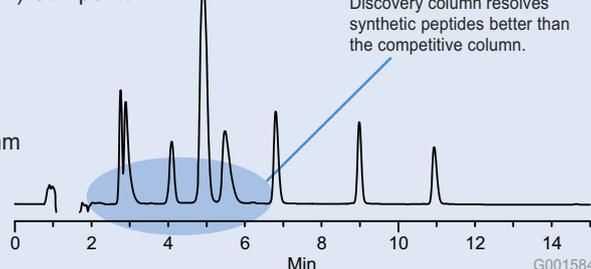
Injection: 10µL, ~0.25µg each peptide (Sigma Peptide Mix, Cat. No. P 2693 containing Arg⁸-vasopressin, bradykinin (fragment 1-5), oxytocin, luteinizing hormone releasing hormone, Met-enkephalin, bradykinin, Leu-enkephalin, bombesin, Substance P) in 0.1%TFA. See sequence in Figure 14.

Gradient: 0-100%B in 14 min after 1 minute delay

(A) Discovery BIO Wide Pore C18



(B) Competitor



Suggestions for Choosing a Discovery BIO Wide Pore Column:

APPLICATIONS	BONDED PHASES
Proteins	BIO Wide Pore C5
Hydrophobic peptides or proteins (e.g. membrane proteins)	BIO Wide Pore C5
Peptide mapping	BIO Wide Pore C18
Proteomics	BIO Wide Pore C18
Scouting	BIO Wide Pore C8 (because of its intermediate hydrophobicity between a C18 and C5)
APPLICATION	SILICA PARTICLE SIZES
LC/MS	3 micron or 5 micron
Fast analysis, or highthroughput applications	3 micron
Peptide mapping	3 micron or 5 micron
Analytical HPLC	3 micron or 5 micron
Preparative	10 micron
APPLICATION	COLUMN ID
LC/MS	2.1mm or smaller
Peptide mapping	4.6mm, 4.0mm, 2.1mm
Analytical HPLC	4.0mm, 4.6mm
Preparative	10mm, 21.2mm
Low level detection or limited sample volume	0.32mm, 0.5mm, 1.0mm

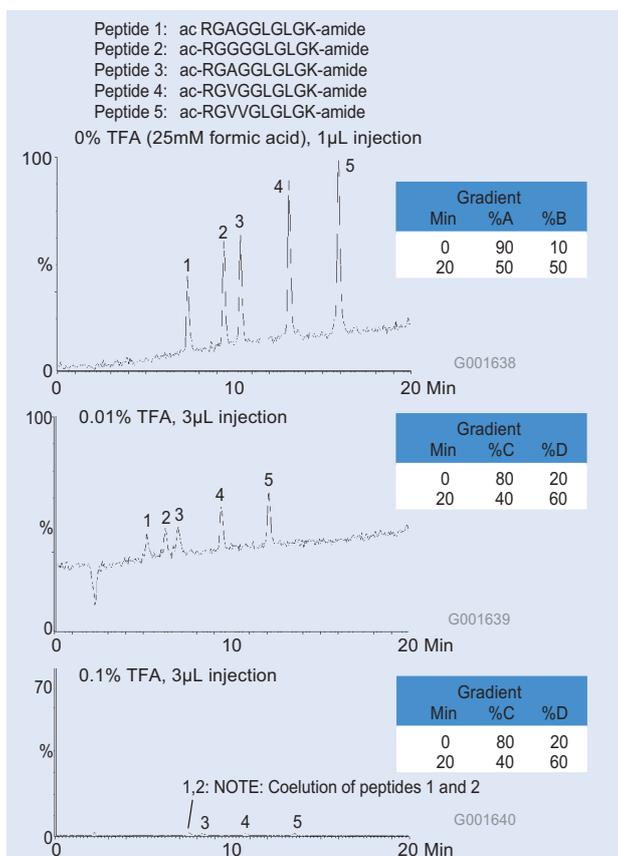
HPLC: Biopolymers

Discovery BIO Wide Pore C18 Columns

Discovery BIO Wide Pore C18



Peptide maps generated by RP-HPLC provide valuable information about protein structure, stability, and purity. To be effective, the RP-HPLC column must be able to resolve a high percentage of the peptides in the sample. The more peptides, the better the information. Discovery BIO Wide Pore C18 gives unsurpassed RP-HPLC resolution of peptide maps from tryptic digests. The improvements in silica and bonded phase chemistry we've incorporated into the Discovery BIO Wide Pore line improve resolution by increasing efficiency and reducing the peak tailing. An added benefit to this is the ability to analyze peptides without TFA in the mobile phase thereby increasing the LC/MS signal.



Effect of Chromatographic Conditions on MS Signals of Peptides

Column: Discovery BIO Wide Pore C18, 15cm x 2.1mm, 3µm
 Cat. No.: 567202-U
 Mobile Phase A: 25mM formic acid in water
 Mobile Phase B: 50:50 (25mM formic acid in water) : (20mM formic acid in MeCN) ¹
 Mobile Phase C: 0.01 or 0.1% TFA in water
 Mobile Phase D: 50:50 (0.01 or 0.1% TFA in water) : (0.01 or 0.1% TFA in MeCN)
 Flow Rate: 0.208mL/min²
 Det.: +ES
 Temp.: ambient
 Inj.: 1µL or 3µL
 Sample: RP Peptide Performance Standard, p/n RPS-P0010 (Alberta Peptide Institute)

1. molarity of formic acid adjusted to provide minimum baseline drift.
2. linear velocity equal to 1mL/min on 4.6mm ID columns.

PARTICLE SIZE	LENGTH x ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY BIO WIDE PORE C18 (300Å, 9.2% CARBON)			
3µm	5 x 0.32	65526-U	
3µm	10 x 0.32	65527-U	
3µm	5 x 0.5	65517-U	
3µm	10 x 0.5	65518-U	
3µm	5 x 1.0	65504-U	
3µm	10 x 1.0	65506-U	
3µm	5 x 2.1	567200-U	
3µm	10 x 2.1	567201-U	
3µm	15 x 2.1	567202-U	
3µm	5 x 4.6	567203-U	
3µm	10 x 4.6	567204-U	
3µm	15 x 4.6	567205-U	
5µm	15 x 0.32	65529-U	
5µm	15 x 0.5	65519-U	
5µm	15 x 1.0	65508-U	
5µm	25 x 1.0	65509-U	
5µm	5 x 2.1	568200-U	
5µm	10 x 2.1	568201-U	
5µm	15 x 2.1	568202-U	
5µm	25 x 2.1	568203-U	
5µm	5 x 4.0	568210-U	
5µm	10 x 4.0	568211-U	
5µm	15 x 4.0	568212-U	
5µm	25 x 4.0	568213-U	
5µm	5 x 4.6	568220-U	
5µm	10 x 4.6	568221-U	
5µm	15 x 4.6	568222-U	
5µm	25 x 4.6	568223-U	
5µm	25 x 10.0	568230-U	
10µm	25 x 4.6	567206-U	
10µm	5 x 10.0	567207-U	
10µm	15 x 10.0	567208-U	
10µm	25 x 10.0	567209-U	
10µm	5 x 21.2	567210-U	
10µm	15 x 21.2	567211-U	
10µm	25 x 21.2	567212-U	
Supelguard Cartridges			
3µm	2 x 2.1 (2/pk)	567270-U	
3µm	2 x 2.1 kit **	567271-U	
3µm	2 x 4.0 (2/pk)	567272-U	
3µm	2 x 4.0 kit **	567273-U	
5µm	2 x 2.1 (2/pk)	568270-U	
5µm	2 x 2.1 kit **	568271-U	
5µm	2 x 4.0 (2/pk)	568272-U	
5µm	2 x 4.0 kit **	568273-U	
10µm	1 x 10.0	567282-U	

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

PROPERTIES:

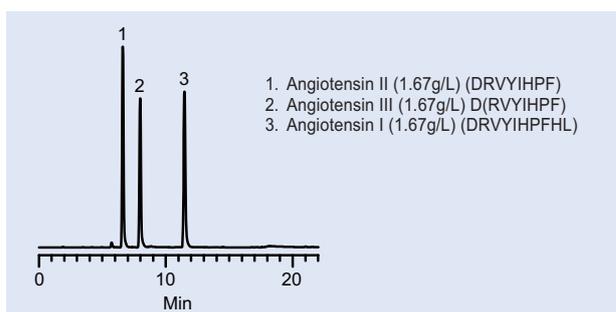
Bonded Phase: Covalently-bonded octadecylsilane, endcapped
 Silica: Spherical, high purity (<10ppm metals)
 Particle Size: 3, 5, and 10mm
 Pore Size: 300Å
 Surface Area: 100m²/g
 %C: ~9%
 Coverage: ~4 µmoles/m²

HPLC: Biopolymers

Discovery BIO Wide Pore C8 Columns

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY BIO WIDE PORE C8 (300Å, 5.0% CARBON)			
3µm	5 x 2.1	567213-U	
3µm	10 x 2.1	567214-U	
3µm	15 x 2.1	567215-U	
3µm	5 x 4.6	567216-U	
3µm	10 x 4.6	567217-U	
3µm	15 x 4.6	567218-U	
Supelguard Cartridges			
3µm	2 x 2.1 (2/pk)	567274-U	
3µm	2 x 2.1 kit **	567275-U	
3µm	2 x 4.0 (2/pk)	567276-U	
3µm	2 x 4.0 kit **	567277-U	
5µm	2 x 2.1 (2/pk)	568370-U	
5µm	2 x 2.1 kit **	568371-U	
5µm	2 x 4.0 (2/pk)	568372-U	
5µm	2 x 4.0 kit **	568373-U	
10µm	1 x 10.0	567284-U	

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.



Resolution of Angiotensins at Neutral pH

Column: Discovery BIO Wide Pore C8, 15cm x 4.6mm, 5µm
Cat. No.: 568322-U
Mobile Phase: (A) 10mM NH₄H₂PO₄/NH₄OH, pH 7;
(B) 50:50, (20mM NH₄H₂PO₄/NH₄OH, pH 7):MeCN
Flow Rate: 1mL/min
Temp: 30°C
Det.: 215nm
Inj.: 6µL in water
Gradient: 30-60% B in 15 min

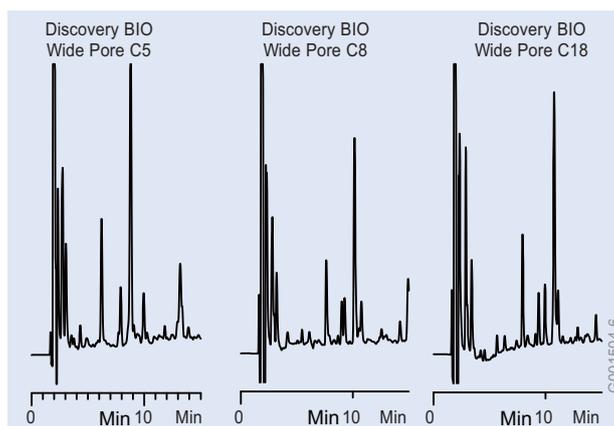
Discovery BIO Wide Pore C8



Discovery BIO Wide Pore C8 is an RP-HPLC phase for proteins and peptides that has hydrophobicity intermediate between the Discovery BIO Wide Pore C5 and the Discovery BIO Wide Pore C18. The difference in hydrophobicity gives it unique selectivity relative to these other phases. It is ideal for peptide mapping because it provides complementary information compared to a C18 separation. Because of its intermediate hydrophobicity we also recommend it for method development or scouting work. As with all Discovery BIO Wide Pore phases, the C8 phase gives efficient, symmetrical peaks, exceptional stability, long column lifetime, and LC/MS compatibility.

PROPERTIES:

Bonded Phase: Covalently-bonded octylsilane, endcapped
Silica: Spherical, high purity (<10ppm metals)
Particle Size: 3, 5, and 10mm
Pore Size: 300Å
Surface Area: 100m²/g
%C: ~5
Coverage: ~4 µmoles/m²



Each Discovery BIO Wide Pore Phase Gives Unique Elution Profiles of Carboxymethylated Apohemoglobin Peptide Fragments

Columns: (A) Discovery BIO Wide Pore C5 (Cat. No. 568422-U),
(B) Discovery BIO Wide Pore C8 (Cat. No. 568322-U),
or (C) Discovery BIO Wide Pore C18 (568222-U),
each 15cm x 4.6mm, 5µm
Mobile Phase: (A) 95:5, (0.1% TFA in water):(0.1% TFA in CH₃CN);
(B) 50:50, (0.1% TFA in water):(0.1% TFA in CH₃CN)
Flow Rate: 1.0mL/min
Temp: 30°C
Detection: 215nm
Injection: 50µL carboxymethylated apohemoglobin tryptic digest in 50mM NH₄HCO₃
Gradient: 0-100%B in 65 min

HPLC: Biopolymers

Discovery BIO Wide Pore C5 Columns

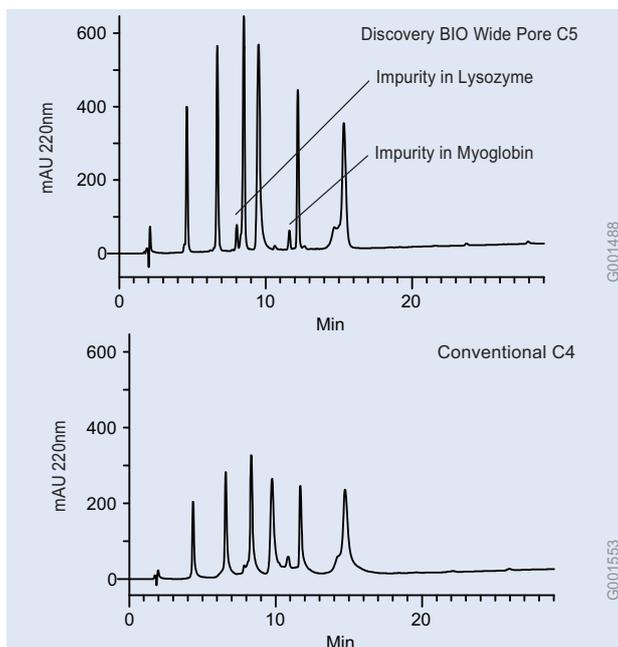
Discovery BIO Wide Pore C5



Discovery BIO Wide Pore C5 was designed for the efficient and reliable separation of proteins and peptides, especially hydrophobic peptides, by RP-HPLC. Long-chain phases, like C8 or C18, are often too hydrophobic for proteins and can cause excessively long retention time or even irreversible binding to the column. For this reason short-chain phases, typically C3 or C4, are often used for RP-HPLC of proteins. However, these short-chain phases are susceptible to hydrolysis resulting in short column lifetime, especially at low pH. The Discovery BIO Wide Pore C5 gives elution order similar to a conventional C4, yet has enhanced pH stability for longer column lifetime. Generally, higher efficiency separations are achievable on the Discovery BIO Wide Pore C5 because of the improvements we have made to the silica and bonded phase chemistry.

PROPERTIES:

Bonded Phase: Covalently-bonded pentylsilane, endcapped
 Silica: Spherical, high purity (<10ppm metals)
 Particle Size: 3, 5, and 10µm
 Pore Size: 300Å
 Surface Area: 100m²/g
 %C: ~3%
 Coverage: ~4 µmoles/m²



Excellent Performance for Proteins

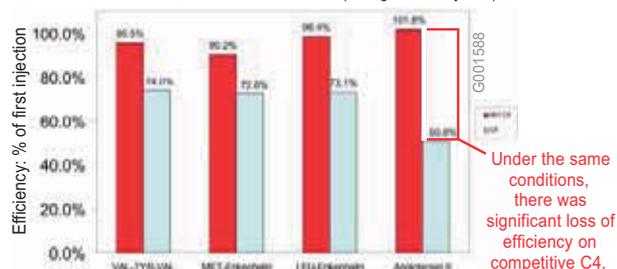
Column: Discovery BIO Wide Pore C5, 15cm x 4.6mm, 5µm
 Cat. No.: 568422-U
 Mobile Phase: (A) 0.1% (v/v) TFA in H₂O : MeCN (75:25),
 (B) 0.1% (v/v) TFA in H₂O : MeCN (25:75)
 Flow Rate: 1mL/min
 Temp: 30°C
 Det.: 220nm
 Inj.: 10µL
 Sample: Protein Test Mix in 0.1% TFA
 Gradient: 0-100% B in 25 min.

PARTICLE SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
DISCOVERY BIO WIDE PORE C5 (300Å, 3.5% CARBON)			
3µm	5 x 0.32	65531-U	
3µm	10 x 0.32	65532-U	
3µm	5 x 0.5	65520-U	
3µm	10 x 0.5	65521-U	
3µm	5 x 1.0	65511-U	
3µm	10 x 1.0	65512-U	
3µm	5 x 2.1	567226-U	
3µm	10 x 2.1	567227-U	
3µm	15 x 2.1	567228-U	
3µm	5 x 4.6	567229-U	
3µm	10 x 4.6	567230-U	
3µm	15 x 4.6	567231-U	
5µm	15 x 0.32	65533-U	
5µm	15 x 0.5	65522-U	
5µm	15 x 1.0	65513-U	
5µm	5 x 2.1	568400-U	
5µm	10 x 2.1	568401-U	
5µm	15 x 2.1	568402-U	
5µm	25 x 2.1	568403-U	
5µm	5 x 4.0	568410-U	
5µm	10 x 4.0	568411-U	
5µm	15 x 4.0	568412-U	
5µm	25 x 4.0	568413-U	
5µm	5 x 4.6	568420-U	
5µm	10 x 4.6	568421-U	
5µm	15 x 4.6	568422-U	
5µm	25 x 4.6	568423-U	
5µm	25 x 10.0	568430-U	
10µm	25 x 4.6	567232-U	
10µm	5 x 10.0	567233-U	
10µm	15 x 10.0	567234-U	
10µm	25 x 10.0	567235-U	
10µm	5 x 21.2	567236-U	
10µm	15 x 21.2	567237-U	
10µm	25 x 21.2	567238-U	
Supelguard Cartridges			
3µm	2 x 2.1 (2/pk)	567278-U	
3µm	2 x 2.1 kit **	567279-U	
3µm	2 x 4.0 (2/pk)	567280-U	
3µm	2 x 4.0 kit **	567281-U	
5µm	2 x 2.1 (2/pk)	568470-U	
5µm	2 x 2.1 kit **	568471-U	
5µm	2 x 4.0 (2/pk)	568472-U	
5µm	2 x 4.0 kit **	568473-U	
10µm	1 x 10.0	567286-U	

** Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

Increased Stability of Discovery BIO Wide Pore C5 over Conventional C4

Efficiency on Discovery BIO Wide Pore C5 is stable even after 25,000 column volumes (222 gradient cycles).



HPLC: Biopolymers

Ion Exchange Chromatography

MCI GEL Ion Exchange Columns

In ion exchange purifications and analyses of biopolymers, ProtEx-DEAE columns (diethylaminoethyl functionality) and ProtEx-SP columns (sulfopropyl functionality) offer significant benefits:

- Excellent separations of protein isoforms
- High resolution at low sample load
- Quantitative recovery – a hydrophilic surface eliminates protein adsorption
- High efficiency (plate number)

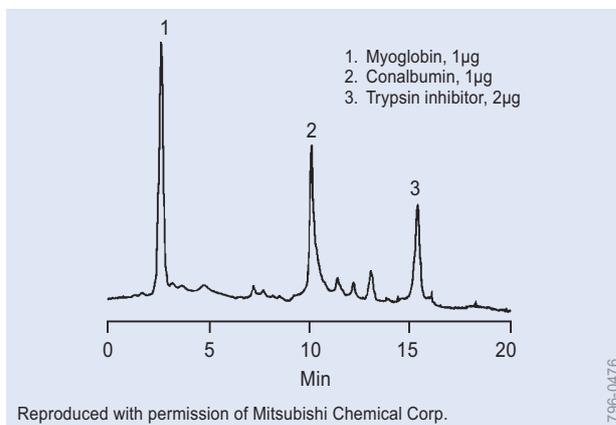
ProtEx columns are 5.0cm x 4.6mm ID polyetheretherketone (PEEK) hardware packed with high performance, monodisperse, 5µm, 1000Å macroporous polymethacrylate beads with a chemically bonded, crosslinked hydrophilic surface. Physical characteristics of these columns are summarized below.

We especially recommend ProtEx columns for applications that require high resolution, such as separating protein isoforms, isozymes, or DNA.

COLUMN CHARACTERISTICS

Dimensions:	5.0cm x 4.6mm ID
Bed Volume:	0.83mL
Flow Rate:	0.5-1.5mL/min
Maximum Backpressure:	735psi (4.9MPa)
Temperature:	4°C - 50°C
pH:	2 - 11
Maximum Loading Capacity:	10mg/injection

Proteins at Low Loading Concentrations



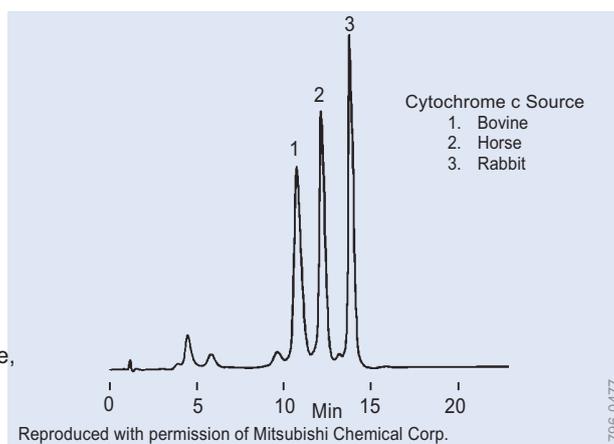
Column: ProtEx-DEAE, 5.0cm x 4.6mm ID, 5µm particles
 Cat. No.: 54742
 Mobile Phase: A = 20mM Tris HCl, pH 8.2
 B = A + 0.5M NaCl
 0% B – 100% B in 20 min
 Flow Rate: 0.5mL/min
 Det.: UV, 280nm

RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No.	Subject
T496177	ProtEx ion exchange HPLC columns
T194882	Mobile phases for ion exchange and chromatofocusing
T495047	Mono Q and Mono S columns

Cytochrome c Species Variants by Ion Exchange HPLC



Column: ProtEx-SP, 5.0cm x 4.6mm ID, 5µm particles
 Cat. No.: 54740-U
 Mobile Phase: A = 20mM Bis-Tris HCl, pH 7.0; B = A + 0.5M NaCl
 24% B – 69% B in 20 min
 Flow Rate: 0.5mL/min
 Det.: UV, 280nm
 Inj.: 10µg each isoform



DESCRIPTION	PART. SIZE	LENGTH x ID (cm x mm)	CAT. NO.	PRICE
ProtEx-SP	5µm	5 x 4.6	54740-U	
ProtEx-DEAE	5µm	5 x 4.6	54742	

Amersham Biosciences Ion Exchange Columns

Mono columns are highly efficient, pH-stable columns designed for high performance ion exchange separations of proteins, peptides, and polynucleotides, in applications including peptide mapping and monoclonal antibody purification. The unique properties of these columns are based on MonoBeads support – a beaded hydrophilic material with the narrowest particle size distribution of any chromatographic support. This monodispersity permits high flow rates at relatively low backpressures.

DESCRIPTION	QTY.	CAT. NO.	PRICE
AMERSHAM BIOSCIENCES ION EXCHANGE COLUMNS			
Mono Q HR5/5	1	54807	
Mono S HR5/5	1	54808	
Mono P HR5/5	1	54809	
CHROMATOFOCUSING REAGENTS			
Pharmalyte 8-10.5	25mL	P2147-25ML	
Polybuffer 74	100mL	P9652-100ML	
Polybuffer 74	250mL	P9652-250ML	
Polybuffer 96	25mL	P9777-25ML	
Polybuffer 96	100mL	P9777-100ML	
Polybuffer 96	250mL	P9777-250ML	

HPLC: Biopolymers Ion Exchange Chromatography

TSK-GEL Ion Exchange Columns

TSK-GEL ion exchange columns are highly efficient and combine sample purification with excellent recovery rates. Anionic and cation exchangers are available on porous polymer-based and silica-based matrices, and in nonporous resin (NPR) columns. The various column types are described in the table below.

TSK-GEL 5PW and NPR ion exchange columns are stable between pH 2-7.5. TSK-GEL SW columns can be used from pH 2-7.5. TSK-GEL 2SW ion exchange columns (125Å pores) are best suited for small molecular weight solutes, such as nucleotides. Larger biomolecules, including peptides and small proteins, can be analyzed on TSK-GEL 3SW ion exchange columns (250Å pores). The wide-pore (1000Å), polymer-based 5PW columns are suitable for analyses and purifications of large proteins and nucleic acids. Sample capacity for a 7.5cm x 7.5mm 5PW ion exchange column is approximately 1mg.

Proteins and nucleic acids can be analyzed 3-5 times faster on a nonporous TSK-GEL NPR column. The sample capacity of these columns for proteins is, however, 50-100 times smaller. TSK-GEL DEAE-NPR columns are commonly used to separate DNA fragments, particularly those obtained from the polymerase chain reaction (PCR). We strongly recommend using a DEAE-NPR guard column to protect the analytical column when analyzing PCR fragments. SP-NPR columns can provide fast results in hemoglobin A1c screening. Due to their small particle size (2.5µm), packings in TSK-GEL NPR columns must be protected by using a pre-column filter containing a 0.5µm frit (Rheodyne in-line filter or Sigma-Aldrich pre-column filter).

CHARACTERISTIC	DEAE-5PW	DEAE-3SW	DEAE-2SW	DEAE-NPR	
ANION EXCHANGE COLUMNS					
Matrix	hydrophilic resin	silica	silica	hydrophilic resin	
Particle Size (µm)	10	10	5	2.5	
Pore Size (Å)	1000	250	125	nonporous	
Functional Group	-CH ₂ CH ₂ N ⁺ (C ₂ H ₅) ₂	-CH ₂ CH ₂ N ⁺ (C ₂ H ₅) ₂	-CH ₂ CH ₂ N ⁺ (C ₂ H ₅) ₂	-CH ₂ CH ₂ N ⁺ (C ₂ H ₅) ₂	
Counter Ion	Cl ⁻	Cl ⁻	Cl ⁻	Cl ⁻	
pH Range	2 - 12	2 - 7.5	2 - 7.5	2 - 12	
Exclusion Limit (PEG, Dalton)	1,000,000	30,000	10,000	500	
Capacity (mg BSA/mL)	30	120	not available	5	
Small Ion Capacity	>0.1meq/mL	>0.3meq/g	>0.3meq/mL	>0.15meq/mL	
pKa	11.2	11.2	11.2	11.2	
CHARACTERISTIC	SP-5PW	SP-NPR	CM-5PW	CM-2SW	CM-3SW
CATION EXCHANGE COLUMNS					
Matrix	hydrophilic resin	hydrophilic resin	hydrophilic resin	silica	silica
Particle Size (µm)	10	2.5	10	5	10
Pore Size (Å)	1000	nonporous	1000	125	250
Functional Group	-CH ₂ CH ₂ CH ₂ SO ₃ ⁻	-CH ₂ CH ₂ CH ₂ SO ₃ ⁻	-CH ₂ COO ⁻	-CH ₂ COO ⁻	-CH ₂ COO ⁻
Counter Ion	Na ⁺	Na ⁺	Na ⁺	Na ⁺	Na ⁺
pH Range	2 - 12	2 - 12	2 - 12	2 - 7.5	2 - 7.5
Exclusion Limit (PEG, Dalton)	1,000,000	500	1,000,000	10,000	30,000
Capacity (mg Hb/mL)	40	5	45	—	110
Small Ion Capacity	>0.1meq/mL	>0.1meq/g	>0.1meq/mL	>0.3meq/mL	>0.3meq/mL
pKa	2.3	2.3	4.2	4.2	4.2

TSK-GEL Ion Exchange (Anion) Columns

DESCRIPTION	PART. SIZE	LENGTH x ID (cm x mm)	CAT. NO.	PRICE
TSK-GEL ION EXCHANGE (ANION) COLUMNS				
DEAE-NPR	2.5µm	3.5 x 4.6	813075	
DEAE-5PW	10µm	7.5 x 7.5	807164	
DEAE-2SW	5µm	25 x 4.6	807168	
Guard Columns and Kits				
DEAE-NPR	5µm	0.5 x 4.6	817088	
DEAE-5PW	20µm	1 x 6.0 kit	807210	
DEAE-SW	10µm	1 x 6.0 kit	807648	

TSK-GEL Ion Exchange (Cation) Columns

DESCRIPTION	PART. SIZE	LENGTH x ID (cm x mm)	CAT. NO.	PRICE
TSK-GEL ION EXCHANGE (CATION) COLUMNS				
CM-5PW	10µm	7.5 x 7.5	813068	
SP-NPR	2.5µm	3.5 x 4.6	813076	
SP-5PW	10µm	7.5 x 7.5	807161	
CM-2SW	5µm	25 x 4.6	807167	
CM-3SW	10µm	7.5 x 7.5	807162	
Guard Column Kits				
CM-5PW ¹	20µm	1 x 6.0 kit	813069	
SP-5PW	20µm	1 x 6.0 kit	807211	
CM-SW ¹	10µm	1 x 6.0 kit	807650	

¹ Kits include one cartridge, a stand-alone holder, 5mL packing, 5cm of 1/16" tubing, and 2 nuts and ferrules.

RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No.	Subject
T494077	TSK-GEL Ion Exchange Columns
T109862	TSK-GEL NPR Columns

HPLC: Biopolymers

Gel Filtration Chromatography

Amersham Biosciences Gel Filtration Columns

For use in an HPLC system, use unions described in Accessories for HR and HiLoad Columns

Fast Desalting Column -Ready for use in an FPLC system.

Superdex HR Column -Filter Kit HR 10, a filter tool, wrench, and instructions are included with Superdex HR columns.

Hi-Load Superdex Column -A dismantling tool, support screen, 10mm net ring, O-ring, and domed nut (in an accessory bag) are included.

DESCRIPTION	EXCLUSION LIMIT	SEPARATION RANGE (PROTEINS)
Fast Desalting Column	>5,000 Da	1,000-4,000
Superdex Peptide	>20,000 Da	100-7,000
Superdex 75	>100,000 Da	3,000-70,000
Superdex 200	>300,000 Da	10,000-600,000

DESCRIPTION	QTY.	CAT. NO.	PRICE
FAST DESALTING COLUMNS			
Fast Desalting HR10/10		54804-U	
PD-10 Columns ¹	30	54805	
Empty PD-10 ¹	50	54806	
GEL FILTRATION COLUMNS			
Superdex Peptide HR10/30		504165	
Superdex 75 HR10/30		54800-U	
Superdex 200 HR10/30		54801-U	
HiLoad 16/60 Superdex 75 PG		54802-U	
ACCESSORIES FOR HR AND HILOAD COLUMNS			
Filters, HR 5	10	54860-U	
Filter Tool	1	54863	
Unions, 1/16", female to M6 female			
Waters	2	54866	
Valco Adapter, male 10-32/female M6	2	54868	
Column End Plugs (Domed Nut) for M6 male	4	54865	
Capillary Tubing, PTFE			
2m x 1.8mm OD x 0.5mm ID		54870	

¹ Syringe barrel sample preparation columns; cannot be connected to any system.

² Amersham Biosciences columns are supplied ready for installation into an FPLC system. To connect these columns to an HPLC system, use one of the three listed unions.

RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No. Subject
T494076 TSK-GEL SW and SW_{XL} Columns

TSK-GEL Gel Filtration Columns (Silica-Based)

TSK-GEL SW and TSK-GEL SW_{XL} columns contain silica-based, hydrophilic bonded phase packings that minimize interaction with proteins. Properties of these popular columns are summarized in the table below. A 30cm TSK-GEL SW_{XL} column and a 60cm TSK-GEL SW column provide similar resolution, but the SW_{XL} column requires half the time. Sample capacity increases in proportion with column length.

Because TSK-GEL SW_{XL} and TSK-GEL SW columns are silica-based, they must be operated within the recommended pH range of 2.5-7.5. Detailed operating conditions are described in the information accompanying the columns. We recommend protecting these columns with the appropriate SW_{XL} or SW guard column.

TSK-GEL QC-PAK columns provide fast, high resolution analyses, especially in quality control applications. The 15cm glass or stainless steel columns are packed with 5µm SW_{XL} materials.

TSK-GEL COLUMN	PARTICLE SIZE (µm)	PORE SIZE (Å)	SAMPLE MW (GLOBULAR PROTEINS)
Super SW2000	4	125	5-150 x 10 ³
G2000SW _{XL}	5	125	5-150 x 10 ³
G2000SW	10	125	5-100 x 10 ³
Super SW3000	4	250	10-500 x 10 ³
G3000SW _{XL}	5	250	10-500 x 10 ³
G3000SW	10	250	10-500 x 10 ³
G4000SW _{XL}	8	450	20-10,000 x 10 ³
G4000SW	13	450	20-10,000 x 10 ³

Mobile Phase: 0.03M NaCl in 0.1M phosphate buffer, pH 7.0

DESCRIPTION	PART. SIZE	LENGTH x ID (cm x mm)	CAT. NO.	PRICE
TSK-GEL GEL FILTRATION COLUMNS				
G2000SW _{XL}	5µm	30 x 7.8	808540	
G3000SW _{XL}	5µm	30 x 7.8	808541	
G4000SW _{XL}	8µm	30 x 7.8	808542	
Super SW2000	4µm	30 x 4.6	818674	
Super SW3000	4µm	30 x 4.6	818675	
G2000SW	10µm	30 x 7.5	805788	
G2000SW	10µm	60 x 7.5	805102	
G3000SW	10µm	30 x 7.5	805789	
G3000SW	10µm	60 x 7.5	805103	
G4000SW	13µm	30 x 7.5	805790	
G4000SW	13µm	60 x 7.5	805104	

TSK-GEL QC-PAK GEL FILTRATION COLUMNS

GFC 200	5µm	15 x 7.8	816215
GFC 300	5µm	15 x 7.8	816049
GFC 300GL ³	5µm	15 x 8.0	816216

GUARD COLUMNS

SW _{XL}	7µm	4 x 6.0	808543
SW	10µm	7.5 x 7.5	805371

³ Glass.

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

HPLC: Biopolymers

Gel Filtration Chromatography

TSK-GEL Gel Filtration Columns (Polymer-Based)

TSK-GEL PW and TSK-GEL PW_{XL} columns are used in high performance gel filtration separations of water-soluble polymers and oligosaccharides. The hydrophilic polymer matrix has excellent chemical and mechanical stability. Although commonly used with aqueous solvents, the polymer is compatible with up to 50% organic solvent.

TSK-GEL GMPW and TSK-GEL GMPW_{XL} columns are mixed bed columns with calibration curves that are linear over a wide range of molecular weights. Because the pore volume of a mixed bed column is the same as that for a narrow pore size column, the slope of the calibration curve is much steeper, which limits resolution. Mixed bed columns are ideal for preliminary investigations, when the molecular weight composition of a sample is unknown. Then, unless the molecular weight distribution of the sample is very broad, one selects a second column (or series of columns) with a pore size (or range of pore sizes) that can provide optimum resolution.

TSK-GEL G-Oligo-PW columns are specially prepared for separating noncharged or cationic oligomers. A small residual positive charge makes G-Oligo-PW and G2000PW columns unsuitable for analyses of anionic oligomers.

DESCRIPTION	PART. SIZE	LENGTH x ID (cm x mm)	CAT. NO.	PRICE
TSK-GEL GEL FILTRATION COLUMNS				
G-Oligo-PW	6µm	30 x 7.8	808031	
G2500PW _{XL}	6µm	30 x 7.8	808020	
G3000PW _{XL}	6µm	30 x 7.8	808021	
G4000PW _{XL}	10µm	30 x 7.8	808022	
G5000PW _{XL}	10µm	30 x 7.8	808023	
G6000PW _{XL}	13µm	30 x 7.8	808024	
GMPW _{XL}	13µm	30 x 7.8	808025	
G-DNA-PW	10µm	30 x 7.8	808032	
G2000PW	10µm	30 x 7.5	805761	
G2500PW	10µm	30 x 7.5	808028	
G3000PW	10µm	30 x 7.5	805762	
G4000PW	17µm	30 x 7.5	805763	
G5000PW	17µm	30 x 7.5	805764	
G6000PW	17µm	30 x 7.5	805765	
GMPW	17µm	30 x 7.5	808026	
GUARD COLUMNS				
G-Oligo-PW	12µm	4 x 6.0	808034	
PW _{XL}	12µm	4 x 6.0	808033	
PW	12µm	7.5 x 7.5	806762	

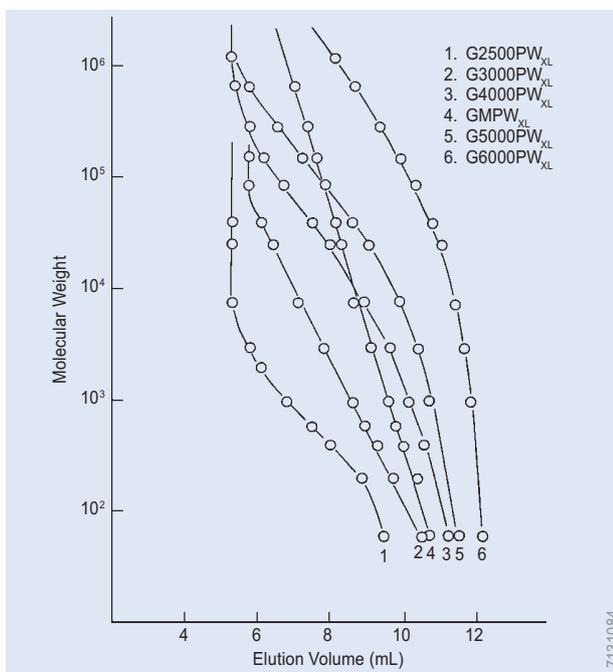
RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No. Subject
T494075 TSK-GEL PW and PW_{XL} Columns

COLUMN	PARTICLE SIZE (µm)	PORE SIZE (Å)	SAMPLE MW	
			PEGs/PEOs	DEXTRANS
G-Oligo-PW	6	125	<2000	—
G2000PW	10	125	<2000	—
G2500PW _{XL}	6	<200	<3000	—
G2500PW	10	<200	<3000	—
G3000PW _{XL}	6	200	<50,000	<60,000
G3000PW	10	200	<50,000	<60,000
G4000PW _{XL}	10	500	2000-300,000	1000-700,000
G4000PW	17	500	2000-300,000	1000-700,000
G5000PW _{XL}	10	1000	4000-1,000,000	50,000-7,000,000
G5000PW	17	1000	4000-1,000,000	50,000-7,000,000
G6000PW _{XL}	13	>1000	40,000-8,000,000	500,000-50,000,000
G6000PW	17	>1000	40,000-8,000,000	500,000-50,000,000
G-DNA-PW	10	4000	40,000-8,000,000	—
GMPW _{XL}	13	<100-1000	500-8,000,000	<50,000,000
GMPW	17	<100-1000	500-8,000,000	<50,000,000

Mobile Phase: Polyethylene glycols/polyethylene oxides - distilled water
Mobile Phase: Dextrans - 0.2M phosphate buffer, pH 6.8



PEG/PEO Calibration Curves on TSK-GEL PW_{XL} Columns

Column 1: G2500PW_{XL}, 30cm x 7.8mm, 6µm particles

Cat. No.: 808020

Column 2: G3000PW_{XL}, 30cm x 7.8mm, 6µm particles

Cat. No.: 808021

Column 3: G4000PW_{XL}, 30cm x 7.8mm, 10µm particles

Cat. No.: 808022

Column 4: GMPW_{XL}, 30cm x 7.8mm, 13µm particles

Cat. No.: 808025

Column 5: G5000PW_{XL}, 30cm x 7.8mm, 10µm particles

Cat. No.: 808023

Column 6: G6000PW_{XL}, 30cm x 7.8mm, 13µm particles

Cat. No.: 808024

Mobile Phase: DI water

Flow Rate: 1mL/min

Det.: refractive index

Inj.: polyethylene glycols and polyethylene oxides

HPLC: Biopolymers

Reversed-Phase, Affinity, Hydrophobic Interaction Chromatography

SUPELCOSIL Wide Pore (300Å) Reversed-Phase Columns

SUPELCOSIL Wide Pore reversed-phase columns continue to be available. Discovery BIO Wide Pore reversed-phase columns provide enhanced performance and are recommended as the first choice. The Discovery BIO Wide Pore columns can be found on the preceding pages.

PARTICLE SIZE	LENGTH x ID (cm x mm)	CAT. NO.	PRICE
SUPELCOSIL LC-318 (300Å, 6.0% CARBON)			
5µm	5 x 4.6	58852	
5µm	25 x 4.6	58858	
SUPELCOSIL LC-318 Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59512	
5µm	2 x 4.0 kit ¹	59502	
SUPELCOSIL LC-308 (300Å, 3.5% CARBON)			
5µm	5 x 4.6	58851	
5µm	25 x 4.6	58857	
SUPELCOSIL LC-308 Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59511-U	
5µm	2 x 4.0 kit ¹	59501	
SUPELCOSIL LC-3DP (300Å, 4.0% CARBON)			
5µm	25 x 4.6	58859	
SUPELCOSIL LC-3DP Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59513	
SUPELCOSIL LC-304 (300Å, 2.7% CARBON)			
5µm	5 x 4.6	58823	
5µm	25 x 4.6	58824	
SUPELCOSIL LC-304 Supelguard Cartridges			
5µm	2 x 4.0 (2/pk)	59592	
5µm	2 x 4.0 kit ¹	59591	
SUPELCOSIL LC-3Si (300Å)			
5µm	25 x 6.2	58965	
SUPELCOSIL LC-Si Supelguard Cartridges (use for LC-3Si)			
5µm	2 x 4.0 (2/pk)	59560	
5µm	2 x 4.0 kit ¹	59550	

RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No.	Subject
T100795	Separating Proteins and Peptides using SUPELCOSIL columns.
T494079	TSK-GEL Reversed-Phase Columns
T494080	TSK-GEL Affinity Columns
T494078	TSK-GEL Hydrophobic Interaction Columns

TSK-GEL Reversed-Phase Columns

DESCRIPTION	PART. SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
SILICA-BASED COLUMNS				
Oligo-DNA RP	5µm	15 x 4.6	813352	
Super ODS	2µm	5 x 4.6	818154	
Super ODS	2µm	10 x 4.6	818197	
Guard Filters				
Filter		(3/pk)	818207	
Stand-Alone Holder for Filters			818206	
RESIN-BASED COLUMNS				
C18-NPR	2.5µm	3.5 x 4.6	814005	
C18-4PW	7µm	15 x 4.6	813351	
Phenyl-5PW RP	10µm	7.5 x 4.6	808043	
Guard Filters				
Filter		(3/pk)	818207	
Stand-Alone Holder for Filters			818206	

TSK-GEL Affinity Columns

DESCRIPTION	PART. SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
ABA-5PW	10µm	7.5 x 7.5	813067	
Boronate-5PW	10µm	7.5 x 7.5	813066	
Chelate-5PW	10µm	7.5 x 7.5	808645	
Guard Column Kits²				
Chelate-5PW	20µm	1 x 6.0 kit	808647	
Heparin-5PW	20µm	1 x 6.0 kit	813121	

TSK-GEL Hydrophobic Interaction (HIC) Columns

DESCRIPTION	PART. SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
Butyl-NPR	2.5µm	3.5 x 4.6	814947	
Ether-5PW	10µm	7.5 x 7.5	808641	
Phenyl-5PW	10µm	7.5 x 7.5	807573	
Guard Column Kits²				
Phenyl-5PW	20µm	1 x 6.0 kit	807652	

¹ Kits include one cartridge, a stand-alone holder, a piece of tubing, and 2 nuts and ferrules.

² Kits include one cartridge, a stand-alone holder, 5mL packing, 5cm of 1/16" tubing, and 2 nuts and ferrules.

HPLC: Industrial Polymers Gel Permeation Chromatography

TSK-GEL Gel Permeation Columns

TSK-GEL H_{HR} gel permeation columns are stable in solvents having a wide range of polarities. The particles do not swell or shrink as the solvent is changed from toluene through methanol. However, these columns cannot be used with polar solvents, such as water or water:methanol mixtures. Spherical $5\mu\text{m}$ polystyrene divinylbenzene particles provide a minimum of 16,000 plates per defined 30cm x 7.8mm ID column. Eight pore sizes are available (Figure A), ranging from an exclusion limit of about 2000 Dalton for G1000 H_{HR} columns to more than 10,000,000 Dalton for G7000 H_{HR} columns. The four mixed bed columns (H, L, M, N) feature extended linear molecular weight operating ranges for sample screening or more formal analyses.

COLUMN	ANALYTE MOLECULAR WEIGHT RANGE (DALTON)
G1000H	<1500
G2000H	<4000
G2500H	< 1.2×10^4
G3000H	< 3.0×10^4
G4000H	< 5.5×10^5
G5000H	< 1.5×10^6
G6000H	< 1×10^7
G7000H	< 5×10^7
GMH-H	< 1×10^7
GMH-L	< 1.0×10^4
GMH-M	< 1.0×10^6

DESCRIPTION	PART. SIZE	LENGTH X ID (cm x mm)	CAT. NO.	PRICE
TSK-GEL H_{HR} GEL PERMEATION COLUMNS				
G1000 H_{HR}	5 μm	30 x 7.8	817352	
G2000 H_{HR}	5 μm	30 x 7.8	817353	
G2500 H_{HR}	5 μm	30 x 7.8	817354	
G3000 H_{HR}	5 μm	30 x 7.8	817355	
G4000 H_{HR}	5 μm	30 x 7.8	817356	
G5000 H_{HR}	5 μm	30 x 7.8	817357	
G7000 H_{HR}	5 μm	30 x 7.8	817359	
GMH H_{HR} -L	5 μm	30 x 7.8	817362	
GMH H_{HR} -M	5 μm	30 x 7.8	817392	
GMH H_{HR} -H	5 μm	30 x 7.8	817360	
Guard Columns				
H_{HR} -L ¹	7 μm	4 x 6.0	817368	
TSK-GEL H_{XL} GEL PERMEATION COLUMNS				
G1000 H_{XL}	5 μm	30 x 7.8	816131	
G2000 H_{XL}	5 μm	30 x 7.8	816134	
G2500 H_{XL}	5 μm	30 x 7.8	816135	
G3000 H_{XL}	6 μm	30 x 7.8	816136	
G4000 H_{XL}	6 μm	30 x 7.8	816137	
G5000 H_{XL}	9 μm	30 x 7.8	816138	
G6000 H_{XL}	9 μm	30 x 7.8	816139	
GMH H_{XL}	9 μm	30 x 7.8	816141	
GMH H_{XL} -HT	13 μm	30 x 7.8	807112	
Guard Columns				
H_{XL} -L ²	6 μm	4 x 6.0	807113	
H_{XL} -H ³	13 μm	4 x 6.0	813727	

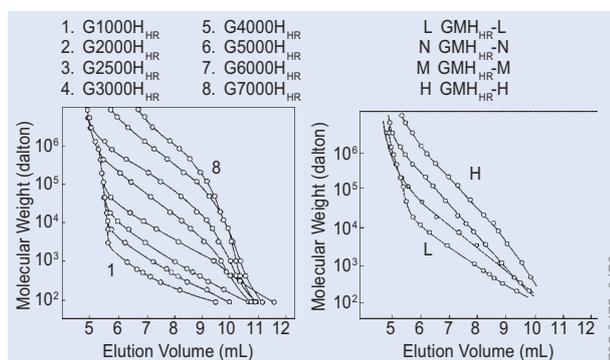
¹ Use to protect G1000 H_{HR} – G4000 H_{HR} and GMH H_{HR} -L columns.

² Use to protect G1000 H_{XL} – G4000 H_{XL} columns.

³ Use to protect G5000 H_{XL} – G6000 H_{XL} columns.

Using Calibration Curves

GPC is widely used for fingerprinting molecular weights of industrial polymers. For compounds of similar molecular shape, a sigmoidal calibration curve is obtained by plotting the logarithm of molecular weight (MW) versus the elution volume. For molecules of known weight, the optimal separation range is defined by the linear portion of this curve. Once a calibration curve is prepared, the elution volume for a polymer of similar shape, but unknown weight, can be used to determine the MW. Results are most accurate when the investigator prepares the calibration curve and determines the molecular weight of the unknown molecule on the same day, with the same mobile phases, etc.



Sample Elution by Molecular Weight

Columns: TSK-GEL H_{HR} , 30cm x 7.8mm ID, 5 μm particles
 Mobile Phase: tetrahydrofuran
 Temp.: ambient
 Flow Rate: 1mL/min
 Det.: UV, 254nm
 Inj.: polystyrene standards

RELATED INFORMATION

Request free literature by phone or fax, or see our website.

No. Subject
 T496085 TSK-GEL H_{HR} Columns

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco

Liquid
Chromatography

SUPELCO

HPLC

Custom, Endfittings, Connectors

Custom Prepared HPLC Columns

If the column of your choice is not listed as a stock product in our catalog, we may be able to prepare it for you as a custom. Below is our current listing of available packings. For the most current listing of hardware, and a price quotation, please contact our Technical Service.

Delivery:

We typically ship custom prepared analytical HPLC columns within 5 to 7 business days to anywhere in the world. Larger sizes (preparative and special requests) may take longer.

Performance Testing:

Supelco's custom prepared columns are fully tested for efficiency and symmetry. Please let us know if you have a special test or

PHASE	PARTICLE SIZE (µm)
DISCOVERY BIO WIDE PORE PHASES	
RP-AmideC16	5
C18	5
C8	5
Cyano	5
DISCOVERY HS PHASES	
C18	3
C18	5
C18	10
F5	3
F5	5
F5	10
PEG	3
PEG	5
PEG	10
DISCOVERY BIO WIDE PORE PHASES	
C18	3
C18	5
C18	10
C8	3
C8	5
C8	10
C5	3
C5	5
C5	10
SUPELCOSIL PHASES	
ABZ ⁺ Plus	3
ABZ ⁺ Plus	5
ABZ ⁺ Plus	12
LC-ABZ	5
Suplex pKb-100	5
LC-18-DB	3
LC-18-DB	5
LC-18	3
LC-18	5
LC-18	12

PHASE	PARTICLE SIZE (µm)
SUPELCOSIL PHASES, CONT'D	
LC-318	5
LC-DABS	3
LC-PAH	3
LC-PAH	5
LC-18-S	5
LC-18-T	3
LC-18-T	5
LC-8-DB	3
LC-8-DB	5
LC-8	3
LC-8	5
LC-8	12
LC-308	5
LC-DP	5
LC-3DP	5
LC-F	5
LC-304	5
LC-CN	3
LC-CN	5
LC-PCN	5
LC-1	5
LC-Si	3
LC-Si	5
LC-Si	12
LC-NH ₂	3
LC-NH ₂	5
LC-NH ₂ -NP	5
LC-Diol	5
LC-3Diol	5
SAX1	5
LC-SCX	5
Hisep	5
SUPELCOGEL PHASES	
TPR-100	5
ODP-50	5
Ion Exclusion	9

Replacement Discovery/SUPELCOSIL Column Endfittings

Use when fittings become damaged or with replacement cartridges. (pk. of 2)



DESCRIPTION	CAT. NO.	PRICE
For 2.1mm columns	55201-U	
For 3.0mm, 4.0mm, 4.6mm columns	55200-U	

Column Connector

Connects two Discovery or SUPELCOSIL analytical columns. Zero dead volume design.



DESCRIPTION	CAT. NO.	PRICE
Column Connector	55213	

Order: 1.800.325.3010 Technical Service: 1.800.359.3041 Web: www.sigma-aldrich.com/supelco