Race through separations with revolutionary technology

Chromolith® HPLC columns

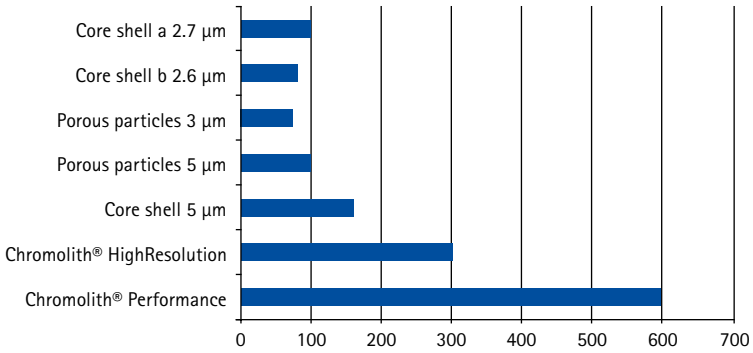
Merck Millipore is a division of MERCK

Tired of congestion?

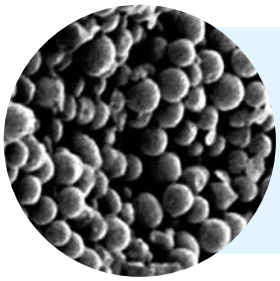
It goes without question that the development of faster separation processes is one of the most important issues in HPLC. Particularly in industry, chromatographers wish to speed up separations, and analyze more samples with the limited financial and human resources available.

One of the main hindrances to speed is congestion. With conventional particle-packed HPLC columns, higher efficiency always comes at the expense of higher back pressure. Even core-shell particle columns, which are designed to lower resistance, still exhibit unacceptable back pressure. Hence, the task is to minimize back pressure in order to maximize speed.

Plates per pressure (N/bar)



All columns are C18e modified, 100-4.6 mm. Sample: anthracene, eluted isocratically using acetonitrile/water (60/40) at 2 mL/min flow rate. Injection volume: 5 µL, detection at 254 nm UV. All analyses performed at room temperature.



A column packed with tiny particles is relatively easy to block since the space between particles is directly proportional to particle size (approx. one sixth of particle diameter). The smaller space leads to higher back pressure and blockage.

SEM image: Silica particles.

Speed up with Chromolith® columns

To truly accelerate chromatographic separations, there's no better choice than Merck Millipore's Chromolith® HPLC columns. Due to their revolutionary monolithic technology, Chromolith® columns provide excellent separations in a fraction of the time required by conventional particulate columns.

The secret to the speed of Chromolith® columns is their exceptionally low back pressure. Produced from a continuous piece of porous silica using a sol-gel process, Chromolith® columns possess a defined bimodal pore structure with macro and mesopores in the micro and nanometer range. The high permeability and porosity of the silica skeleton, and the resulting low back pressure allow for more flexible flow rates than particle-packed columns. As a result, Chromolith® HPLC columns enable high-throughput analysis without loss of separation efficiency or peak capacity.

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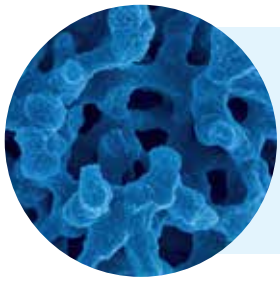

The thin skeleton ensures fast mass transfer, while the large channels allow unobstructed flow through the column.

SEM image: Cross section of a silica monolith
Total porosity > 80 %.

Content

Benefits of Chromolith® HPLC columns

- High flow rates at low pressure
- Flat Van Deemter curve allows flow rate flexibility and flow gradients
- Improved HPLC system security: robustness, reliability, versatility
- Substantially longer column lifetime
- Compatible with various organic solvents, e.g. methanol, ethanol, isopropanol
- Column length no longer pressure limited
- Cost savings from higher sample throughput offset expense of method revalidation in one month
- Suitable for use with standard HPLC instruments
- Available in various sizes and modifications
- Higher porosity allows fast adsorption and desorption kinetics
- Compatible with all mass spectrometers

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Column selection guide

Fields of application

Modifications	Aflatoxins	Alcohols	Aldehydes	Alkaloids	Aliphatic amines	Amino acids	Antibiotics	Aromatic amines	Carboxylic acids	Carotinoids	Explosives	Oils	Esters	Fat soluble vitamins	Lipids	Fatty acids	Flavonoids	Glycols	Ketones	Nitrosamines
Chromolith® **RP-18 endcapped**	1	2	1	1	2	2	1	2		2	1	2	1	1	1	1	1		1	1
Chromolith® **HighResolution RP-18 endcapped**	1	2	1	1	2	2	1	2		2	1	2	1	1	1	1	1		1	1
Chromolith® **RP-8 endcapped**	2		2	2		1	2			2			1			2			2	2
Chromolith® **Phenyl**			2	2			2	1						2			1		2	
Chromolith® **CN**		2										2					1	2		
Chromolith® **DIOL**		2				1				1		2						2		
Chromolith® **Si (silica)**	2									1		1		1	1	1	2	2	2	
Chromolith® **NH_2**		2							1					1						

1 Most commonly used column

2 Column with some successful separation cases

The most suitable column modification can be easily selected using the table above. The first choice is highlighted in dark blue, and indicates the most commonly used column modification for the selected application. The second choice is highlighted in bright blue, and represents a column modification with some successful separation cases reported in scientific literature.

Note: As chromatographic separation depends on many physical and chemical parameters, we cannot guarantee the success of a separation based on the recommended column modification.

1	2			2		2	2	Nucleotides
				1		1	1	PAH
				2	2	1	1	PCB
				1	2	1	1	Peptides
						1	1	Pesticides
			1		2	1	1	Phenols
	2	2				2	2	Phospholipids
			1			1	1	Phthalates
		1		1		1	1	Preservatives
		1			1	2	2	Proteins
1	2		1	2	2	1	1	Steroids
				2	2	1	1	Metabolized steroids
1		1						Sugars
1								Sugar Alcohols
		2	2			1	1	Sulfonamides
1				2		1	1	Sweeteners
1		1				1	1	Water soluble vitamins

Chromolith® columns at a glance

Capillary columns			Analytical columns			Preparative columns	
							
Column internal diameter							
0.05 mm	0.1 mm	0.2 mm	2 mm	3 mm	4.6 mm	10 mm	25 mm
■■■■■■ Page 12	■■■■ ■■■■■■■ ■■■■■■■ Page 12-13	■■■■ ■■■■■■■ Page 12	■■■ ■■■■ ■■■■■ Page 9, 16-17, 35	■■■ ■■■■ ■■■■■ Page 18	■■■ ■■■■ ■■■■■ Page 7-8, 16, 18, 20, 32, 37	■■■■■ Page 30	■■■■■ Page 32
	■■■■■■ Page 12	■■■■■■ Page 12			■■■ ■■■■ ■■■■■ Page 19-22, 37		
	■■■■■■ Page 12				■■■■■ Page 23		
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					■■■■■ Page 26	■■■■■	■■■■■ Page 33
					■■■ ■■■■ ■■■■■ Page 27		
-				Loadability			+
+				Sensitivity			-
+				Solvent saving			-

Chromolith® columns are available in various lengths:

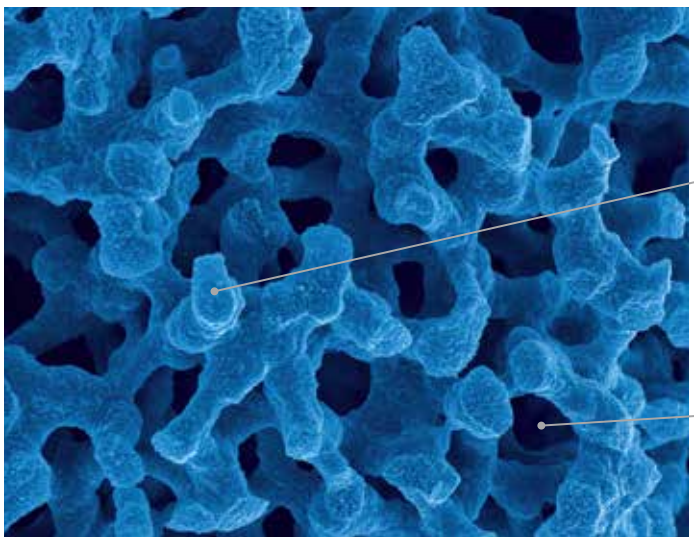
■■■ 25 mm ■■■■ 50 mm ■■■■■ 100 mm ■■■■■■ 150 mm ■■■■■■■ 300 mm

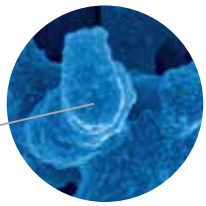
+	Speed	-
-	Resolution	+

Chromolith® HPLC columns
Revolutionary monolithic silica replaces particles

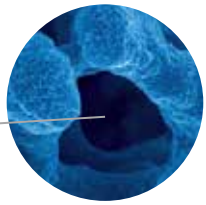
Thanks to their patented monolithic silica technology, Chromolith® HPLC columns allow you to race through separations with maximum robustness and selectivity – at minimal back pressure.

The revolutionary bimodal pore structure of Chromolith® columns provides a unique combination of macropores and mesopores.





Mesopores: Average pore size 13 nm
Form a fine porous structure, and large uniform surface area on which adsorption takes place, thus enabling high-performance chromatographic separation.



Macropores: Average pore size 2 µm
Allow rapid flow of the mobile phase at low back pressure.

SEM image: Cross section of a silica monolith. Total porosity > 80 %.

Characterization of Chromolith® HPLC columns

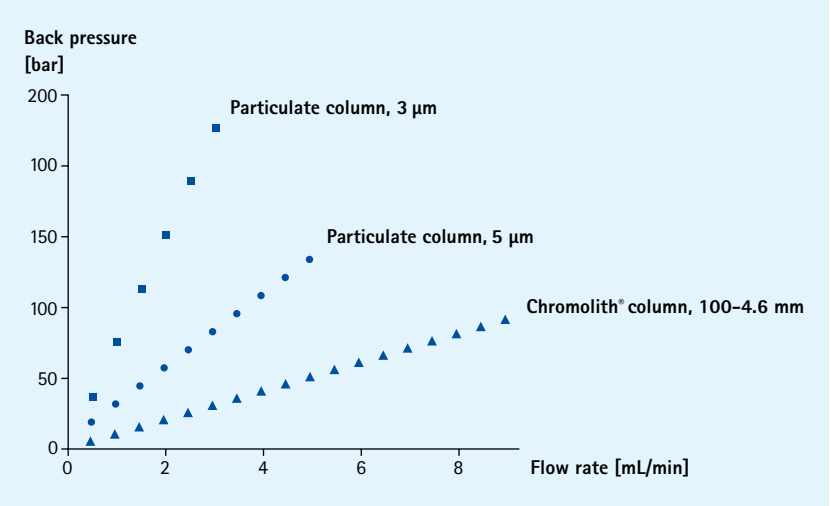
The use of conventional HPLC columns containing 3 or 5 µm silica particles often results in high back pressure. This reduces column lifetime, system robustness and the operational range of flow rates. As a result, these columns are limited in length and in their number of theoretical plates. Attempts have been made to increase plate count by decreasing particle size, but this further raises back pressure, and restricts the variety of separations that can be satisfactorily achieved.

Another means of accelerating chromatographic analysis is through laboratory automation of HPLC systems. This method has come a long way toward improving sample throughput by enabling 24-hour operation. However, the systems are still dependent on the separation technology itself, that is, the separation columns available.

The optimal solution is a column that offers faster throughput without the risk of plugging: the Chromolith® column. In contrast to conventional HPLC columns, Chromolith® columns are not packed with small silica particles. Instead, each column consists of a single rod of high-purity polymeric silica gel with a bimodal pore structure of macro and mesopores. This unique construction enables highly efficient separations at unbeatable speeds.

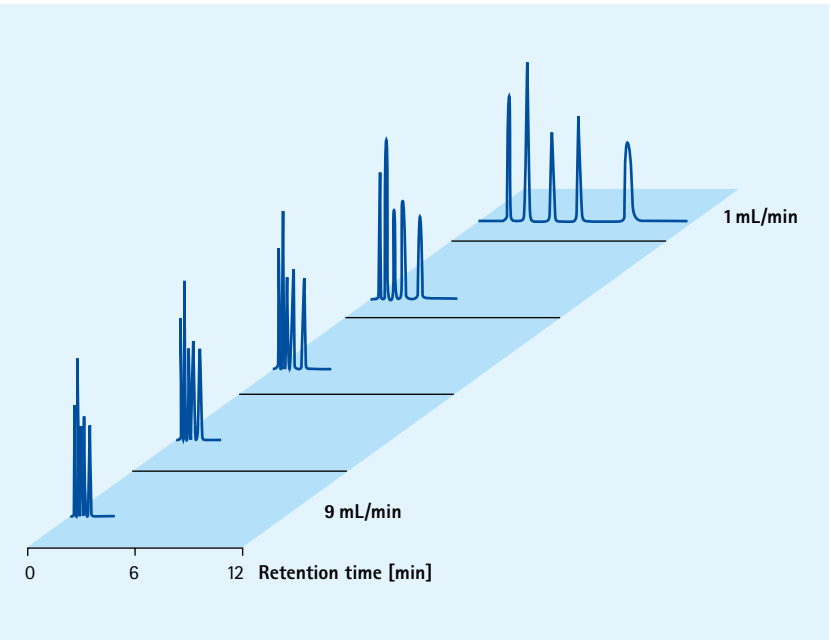
Analysis speed

Chromolith® columns owe their rapid separation speed to their unique bimodal pore structure of macro and mesopores. The **macropores** reduce column back pressure and allow the use of faster flow rates, thereby considerably reducing analysis time.
The **mesopores** form a fine porous structure, which creates a very large active surface area for high-efficiency separations.



Column back pressure at different flow rates. Comparison of a Chromolith® Performance column, 100–4.6 mm vs. equivalent classical particulate HPLC columns.

With Chromolith® columns, flow rates can now easily be varied from 1 mL up to 9 mL per minute with the same high quality resolution.
A mixture of five beta-blocker drugs was analyzed to demonstrate the extreme time savings and high separation efficiency made possible with Chromolith® columns. Due to excellent mass transfer properties of the monolithic skeleton, high-speed separation was possible, even at high flow rates. The beta-blockers were well separated with excellent peak symmetry. At 9 mL/min, analysis time was less than 1 minute, and the column back pressure was only 153 bar.



Chromolith® Performance RP-18e 100–4.6 mm

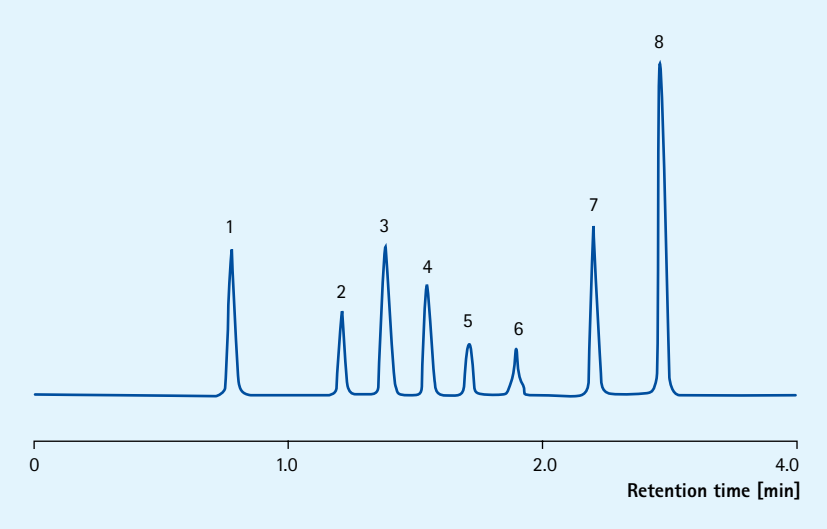
Column	Chromolith® Performance RP-18 endcapped 100–4.6 mm	
Mobile phase	Isocratic acetonitrile / 0.1 % trifluoroacetic acid in water, 20/80 (v/v)	
Pressure	Total pressure (including HPLC system) 25°C	
Detection	UV 220 nm	
Injection volume	5 µL	
Sample	Atenolol	63 µg/mL
	Pindolol	29 µg/mL
	Metoprolol	108 µg/mL
	Celiprolol	104 µg/mL
	Bisoprolol	208 µg/mL

Flow programming

Chromolith® columns respond very quickly to changes in flow rate, giving you maximum flexibility in flow programming. Rates can be adjusted in mid-flow either to enhance the peak definition of the target compound, or to shorten the total separation time once the compound has successfully eluted. This enables clearer separation of two closely eluting peaks, without significantly affecting the total run time. A mid-flow change in rate can also reduce the total run time when certain compounds elute much later than others.

Chromolith® Performance RP-18e 100-4.6 mm

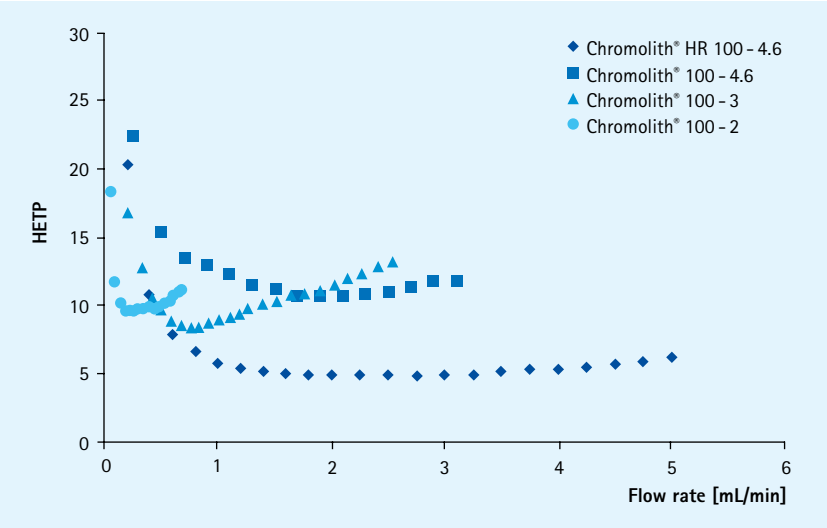
Column	Chromolith® Performance RP-18 endcapped 100-4.6 mm			
Mobile phase	A: Acetonitrile			
	B: 0.1 % Phosphoric acid in water			
Double gradient	**Time**	**% A**	**% B**	**Flow rate**
	0 min	35	65	3 mL/min
	1.8 min	46	54	3 mL/min
	2.2 min	80	20	5 mL/min
	3 min	80	20	5 mL/min
Pressure	90 bar maximum total pressure			
Detection	UV 254 nm			
Temperature	22°C			
Injection volume	10 µL			
Sample	1. Phenol			
	2. 2-Chlorophenol			
	3. 2-Nitrophenol			
	4. 2,4-Dinitrophenol			
	5. Chloro-3-methylphenol			
	6. 2,4-Dinitro-6-methylphenol			
	7. 2,4,6-Trichlorophenol			
	8. Pentachlorphenol			



High separation efficiency

The traditional plate-count method of measuring quality shows that the separation efficiency of Chromolith® columns is better than standard 5 µm particulate columns, and just as good as 3.5 µm columns, but with the ability to continue up to 9 mL/min without reaching HPLC system pressure limits.
The Van Deemter plot of the Chromolith® column clearly demonstrates that separation efficiency does not decrease significantly when flow rate is increased, as is the case with particulate columns. It is therefore possible to operate Chromolith® columns at high flow rates with minimal loss of peak resolution.

For complex separations, it is still necessary to use long columns in order to provide the separation efficiency required for resolution of all compounds of interest. Chromolith® HPLC columns can be connected in series to produce a column with high plate count at low back pressure. (Please see: Chromolith® column coupler). With particulate columns, further column length is prevented by excessive back pressure.



Van Deemter plot of the height equivalent to a theoretical plate (HETP) vs. flow rate for Chromolith® columns.

Long-term stability

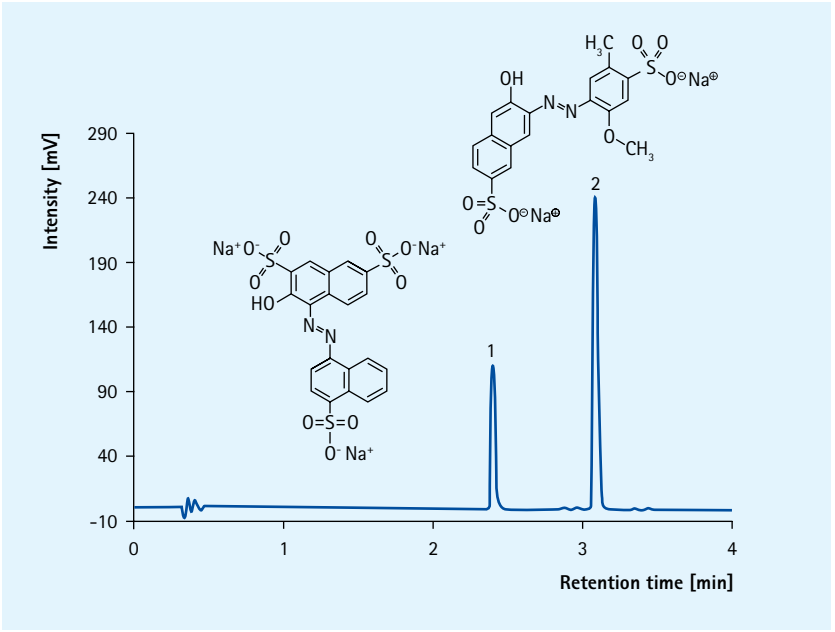
Besides lower back pressure and greater flow rate flexibility, Chromolith® columns also achieve faster equilibration after gradient elution than particle-packed columns of similar dimensions. These features allow high-throughput analysis – without loss of separation efficiency or peak capacity.

Column robustness

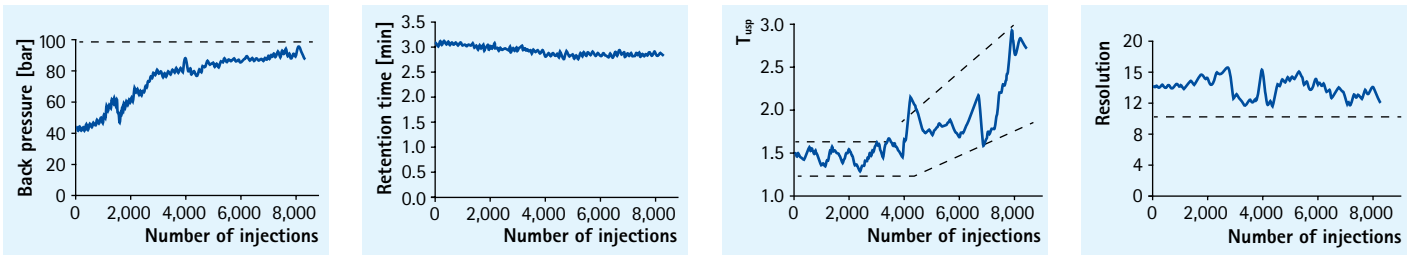
Chromolith® columns offer excellent robustness and unsurpassed column lifetime. This not only ensures maximum reliability and versatility, but also minimizes maintenance on the HPLC system. As a result, Chromolith® columns reduce costs per analysis while enhancing data integrity.

Food colorants in an alcoholic beverage
Chromolith® Performance RP-18e

Column	Chromolith® Performance RP-18 endcapped 50 x 2.0 mm		
Mobile phase (v/v)	A: Acetonitrile B: 0.1 % Phosphoric acid in water		
Gradient Program	**Time**	**% A**	**% B**
	0.00 – 0.50 min	0	100
	0.50 – 4.50 min	0 → 50	100 → 50
	4.50 – 5.00 min	50 → 95	50 → 5
	5.00 – 6.00 min	95	5
	6.00 – 7.00 min	95 → 95	5 → 100
Pressure Drop	45 – 40 bar (648 – 556 psi)		
Detection	UV 500 nm		
Cell	1.4 µL		
Flow Rate	0.4 mL/min		
Temperature	25°C		
Injection volume	2 µL		
Sample	*Prior to analysis, the sample was filtered using a syringe equipped with a 0.45 µm filter disc.*		



Analysis of colorants in rum. Two food colorants in rum, E 123 (Amaranth) and E 129 (Allura Red AC), were analyzed to illustrate the long-term performance and method robustness of Chromolith® columns. More than 8,000 samples (total volume of injected sample: 16 mL + 30 L mobile phase) were analyzed on a 50 x 2.0 mm Chromolith® RP-18 endcapped column.



Evaluation of the robustness of a Chromolith® column for the analysis of food colorants in rum.

The figures above illustrate how column back pressure, peak shape, and the chromatographic resolution between E129 (Allura Red AC) and E123 (Amaranth) are affected with time; 8,300 samples were analyzed. The largest effects are seen on peak shape and back pressure. A similar T_{USP} value is obtained over the first 4,000 injections, after which some deterioration is observed. Despite aging of the column, peak integration and thereby accurate quantitation of the two analytes is achieved. The column back pressure increases with time as sample matrix is accumulated in the column but never reaches over 100 bar (1,450 psi). The chromatographic resolution between the analytes is substantial (Rs >10) with good overall retentivity and no additional disturbing peaks are found in the chromatogram (UV detection at 500 nm).

Capillary columns
0.05 mm / 0.1 mm / 0.2 mm i.d.

Monolithic capillary columns have become increasingly important in the separation of biomolecules, especially in combination with mass spectrometry. In contrast to particulate columns, monolithic capillaries do not require frits, and have a much lower tendency to clog. This allows higher flow rates, improving the speed and quality of biomolecule characterization. To answer the growing interest in micro and nano-HPLC, Merck Millipore offers a wide range of outstanding monolithic silica capillaries with a variety of internal diameters, bonded phases, pore structures, and lengths.

Benefits of capillary columns

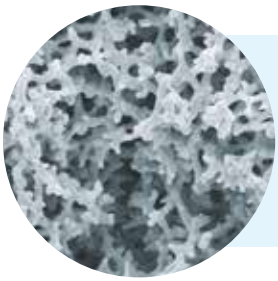
- Higher flow rates than particle-packed capillary columns at low pressure
- Long column lifetime
- Robust and easy handling
- Flow rates from 0.1 – 20 µL/min ensure ideal compatibility with LC/MS systems, with both ESI and APCI interfaces

Chapter content

Chromolith® CapRod®

Chromolith® CapRod® is a capillary column which combines the speed of monolithic silica technology with the sensitivity of nano-LC. This enables superior productivity for high throughput, highly sensitive proteomics-LC applications. Compared to particulate capillary columns, Chromolith® CapRod® capillaries demonstrate better performance with optimal resolution (narrow peak widths), increased productivity (higher sample throughput), and extended column lifetime. Furthermore, column length is less limited than with any other type of column. The capillaries can even be slightly bent to fit any LC configuration or instrument. Chromolith® CapRod® is designed to work with various nano or capillary-LC systems. This provides the highest efficiency and performance when coupled to mass spectrometers, both on-line (ESI, nanospray) and off-line (MALDI).

Compared to classical micro-particulate sorbents, Chromolith® CapRod® can be operated at higher flow rates – without loss of performance, resolution, or limitations due to column back pressure. Separations can be achieved at 1 – 3 µL/min, compared to 200 – 400 nL/min for conventional media on a standard 100 µm LC capillary column. For more complex biological samples, a trapping capillary can be used to protect the separation column, and optimize separation efficiency.



Cross section of the bimodal pore structure of CapRod® with macropores at approx. 2 µm (1 µm for Chromolith® HighResolution columns) and mesopores at 13 nm. The outer diameter of the capillary is 360 nm.

Recommended use and flow rate ranges

Recommended use	**RP-18e** 150 x 0.05 mm	**RP-8e** 150 x 0.1 mm	**RP-18e** 50 x 0.1 mm Trap	**RP-18e** 150 x 0.1 mm	**RP-18e** 300 x 0.1 mm	**RP-18e** 150 x 0.1 mm HR	**RP-18e** 50 x 0.2 mm Trap	**RP-18e** 150 x 0.2 mm	**RP-18e** 150 x 0.2 mm HR
Separation of small molecules	■		■	■	■	■	■	■	■
– of peptides	■	■	■	■	■	■	■	■	■
– of proteins		■							
Micro ESI		■		■	■	■		■	■
Nano ESI	■	■		■	■	■			■
High Resolution						■			■
Flow rates (µL/min)	0.2 – 0.8	0.4 – 3	1 – 10	0.4 – 3	0.2 – 1.5	0.1 – 0.4	10 – 50	5 – 20	0.5 – 2
Max back pressure (bar)	200	200	200	200	200	218	218	218	218

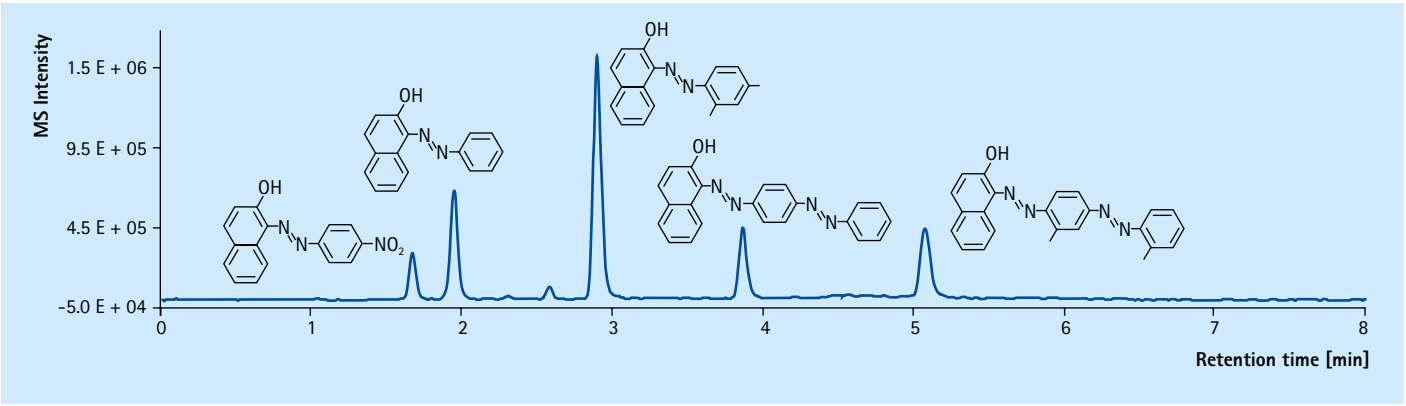
Chromolith® CapRod® analytical capillary columns are supplied complete with sleeves and standard 1/16" PEEK fittings to allow for direct coupling to a UV detector or mass spectrometer.

Separation example: Sudan dyes

Chromolith® CapRod® RP-18e 150-0.1 mm

Column	Chromolith® CapRod® RP-18 endcapped 150 x 0.1 mm
Mobile phase	A: Water + 0.1 % formic acid B: Acetonitrile + 0.1 % formic acid
Gradient	70 % B to 95 % B in 5 min
Flow rate	1.24 µL/min
Pressure Drop	76 bar (1,100 psi)
Detection	nano-ESI(+) 240 – 390 m/z
Temperature	ambient
Diluent	Acetonitrile
Injection volume	2.5 nL

Sample	Compound	Retention Time (min)	Monoisotopic mass (g/mol)	[M+H]+ (m/z)
	1. Para red	1.67	293.08	294.01
	2. Sudan I	1.95	248.09	249.01
	3. Sudan II	2.90	276.13	277.09
	4. Sudan III	3.87	352.13	353.08
	5. Sudan IV	5.08	380.16	381.11

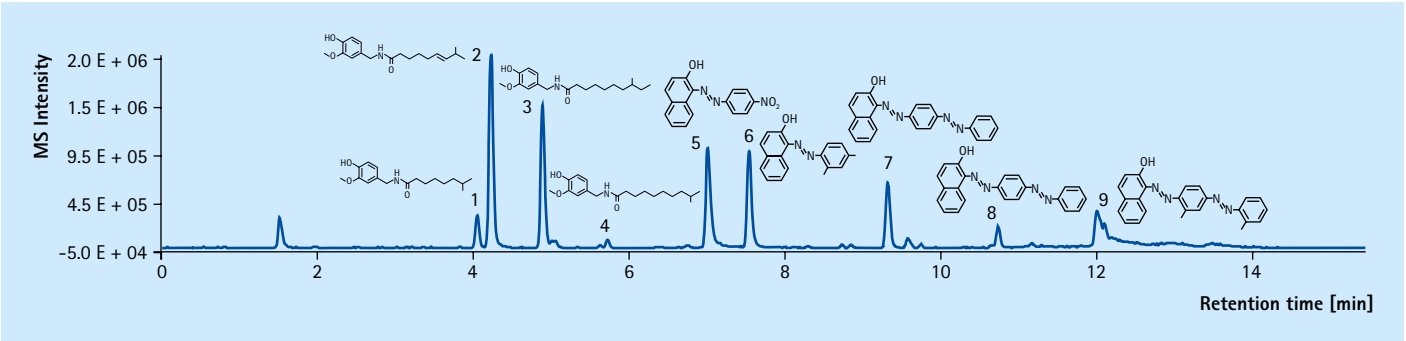


Separation example: Sudan dyes and capsaicinoids in hot chili sauce

Chromolith® CapRod® RP-18e 150-0.1 mm

Column	Chromolith® CapRod® RP-18 endcapped 150 x 0.1 mm
Mobile phase	A: Water + 0.1 % formic acid B: Acetonitrile + 0.1 % formic acid
Gradient	35 % B to 95 % B in 12 min
Flow rate	1.24 µL/min
Pressure Drop	80 bar (1,160 psi)
Detection	nano-ESI(+) 100 – 600 m/z
Temperature	ambient
Diluent	Acetonitrile
Injection volume	2.5 nL

Sample	Compound	Retention Time (min)	Monoisotopic mass (g/mol)	[M+H]+ (m/z)
	1. Nordihydrocapsaicin	4.05	293.20	294.11
	2. Capsaicin	4.23	305.20	306.18
	3. Dihydrocapsaicin	4.90	307.21	308.19
	4. Homodihydrocapsaicin	5.73	321.23	322.11
	5. Para red	7.02	293.08	294.05
	6. Sudan I	7.55	248.09	249.05
	7. Sudan II	9.33	276.13	277.10
	8. Sudan III	10.74	352.13	353.11
	9. Sudan IV	12.11	380.16	381.21



Analytical columns
2 mm / 3 mm / 4.6 mm i.d.

Standard HPLC columns with 3 or 5 µm silica particles often suffer from high back pressure. Hence, they are limited in length, and have a lower number of theoretical plates.

Chromolith® HPLC columns are not packed with small particles. Instead, each column consists of a single monolithic rod of high-purity polymeric silica gel with a revolutionary bimodal pore structure. This allows excellent separations in a fraction of the time that a standard particulate column takes.

Benefits of analytical columns

- Very fast, high-performance results
- Substantially longer column lifetime
- High resistance to column blockage
- Cost savings from higher sample throughput and column durability
- Compatible with all low dead volume LC instruments (UHPLC, UPLC®, HPLC)
- Possibility of flow gradients
- Added column performance by column coupling

Chapter content

Chromolith® RP-18 endcapped

The chemical basis of Chromolith® RP-18 endcapped columns – from starting materials to surface modifications – is the same as high-end particulate columns. Thus, their selectivity is comparable to high-quality C18 endcapped reversed-phase packed columns. This allows the use of standard methods when developing new protocols. The columns are based on high-purity silica, hence they minimize the negative effect of trace metals. Furthermore, they are chemically modified with n-alkyl chains that possess a high ligand density, and fully endcapped to reduce the effect of unmodified silanol groups.

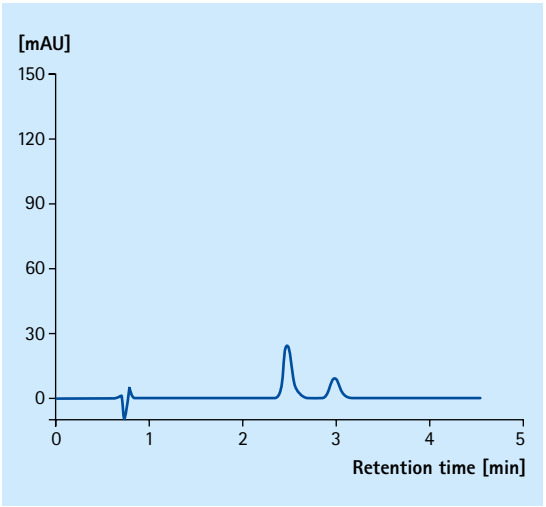
Chromolith® RP-18 endcapped 2 mm i.d. columns: Ultra-high performance on any instrument

Ultra-high performance and extremely low operating pressure make Chromolith® 2 mm columns truly unique. Excellent, ultra-fast results are obtained, not only in the new UHPLC and UPLC® instruments, but equally well in all standard HPLC systems with low dead volume. Chromolith® 2 mm columns have macropores of 1.5 µm in diameter, resulting in a column efficiency that exceeds 100,000 plates/meter. The mesopores are 13 nm (130 A) in diameter, and the surface modification is octadecylsilane with full endcapping.

Increase sensitivity and save solvents with 2 mm i.d. Chromolith® RP-18 endcapped columns

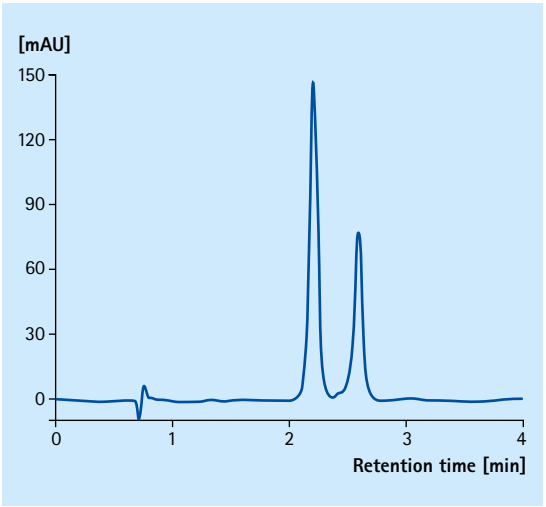
Chromolith® Performance RP-18e 100-4.6 mm

Column	Chromolith® Performance RP-18 endcapped 100-4.6 mm
Mobile phase	A: 100 % Acetonitrile B: 100 % Water + 0.1 % TFA (v/v) C: 100 % Methanol
Isocratic	Initial composition: A/B/C 30/60/10 (v/v/v)
Flow rate	2 mL/min
Pressure	45 bar (4.5 MPa, 65.3 psi)
Detection	Dionex Ultimate 3000 VWD-3400, 2.5 Hz, Response time 0.1 s, UV = 210 nm
Vol. detector cell	11 µL
Temperature	ambient
Injection volume	1 µL
Sample	Bimatoprost Bimatoprost free acid



Chromolith® Performance RP-18e 100-2 mm

Column	Chromolith® Performance RP-18 endcapped 100-2 mm
Mobile phase	A: 100 % Acetonitrile B: 100 % Water + 0.05 % TFA (v/v) C: 100 % Methanol
Isocratic	Initial composition: A/B/C 30/60/10 (v/v/v)
Flow rate	380 µL/min
Pressure	48 bar (4.8 MPa, 70 psi)
Detection	Dionex Ultimate 3000 VWD-3400, 2.5 Hz, Response time 0.1 s, UV = 210 nm
Vol. detector cell	1.4 µL
Temperature	ambient
Injection volume	1 µL
Sample	Bimatoprost Bimatoprost free acid

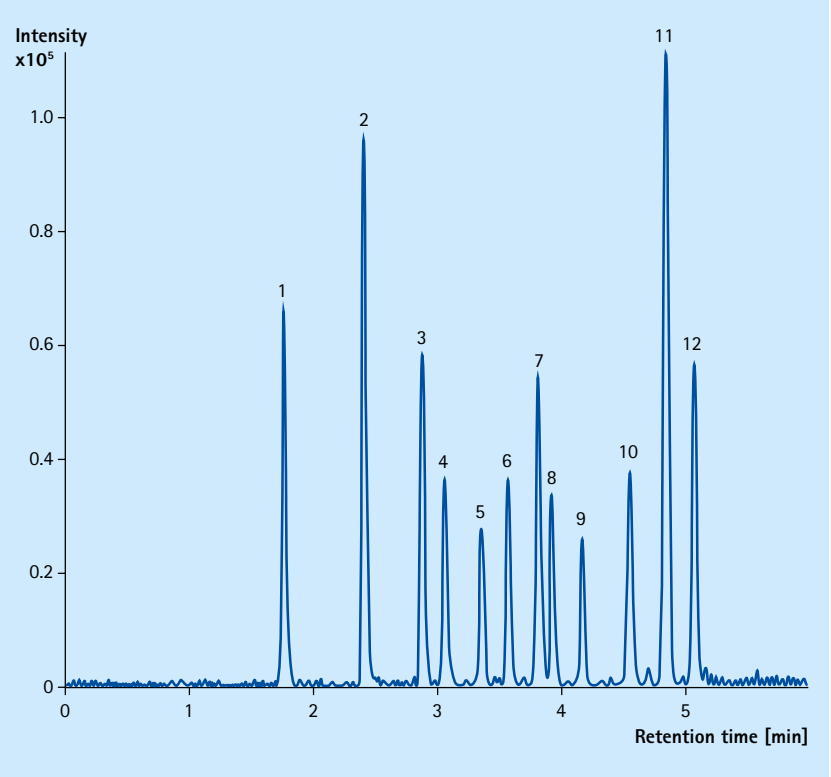


The same separation on a Chromolith® 2 mm i.d. column demonstrates improved sensitivity and solvent savings of 81 %.

Separation of Steroids & Metabolites

Chromolith® FastGradient RP-18e 50-2 mm

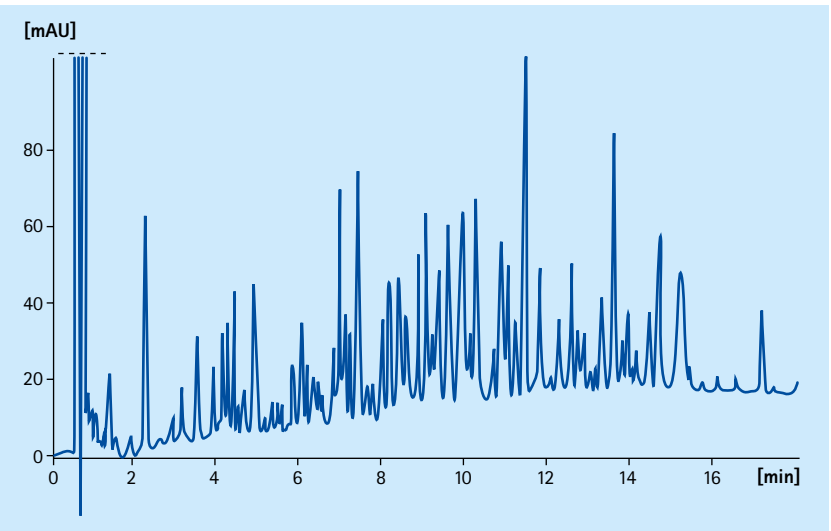
Column	Chromolith® FastGradient RP-18 endcapped 50-2 mm		
Mobile phase	A: ACN + 0.1 % HCOOH B: Water + 0.1 % HCOOH		
Gradient	Time	% A	% B
	0	15	85
	4.5	70	30
	6	70	30
Flow rate	0.5 mL/min		
Pressure	55 – 85 bar		
Detection	MS; Ion Source: ESI; Ion Trap		
Sample	1. Metabolite of Fluoxymesterone		353 m/z
	2. Metabolite of Stanozolol		345 m/z
	3. Metabolite of Danazol		343 m/z
	4. Testosterone		289 m/z
	5. Epitestosterone		289 m/z
	6. Metabolite of Methyltestosterone		271 m/z
	7. Metabolite of Calusterone		285 m/z
	8. Metabolite of Clostebol		305 m/z
	9. Boldenone-acetate		329 m/z
	10. Testosterone-acetate		331 m/z
	11. Nandrolone-17-Propionate		331 m/z
	12. Testosterone-Propionate		345 m/z



Separation example: Proteomics

Chromolith® Performance RP-18e 100-2 mm

Column	Chromolith® Performance RP-18 endcapped 100-2 mm
Mobile phase	A: 95 % H_2O/5 % ACN/0.1 % TFA (v/v/v) B: 5 % H_2O/95 % ACN/0.085 % TFA (v/v/v)
Gradient	from 5 % B to 50 % B in 20 min
Flow rate	0.3 mL/min
Detection	UV 214 nm
Sample	1 µL BSA digest (1 mg/mL)



Chromolith® RP-18 endcapped

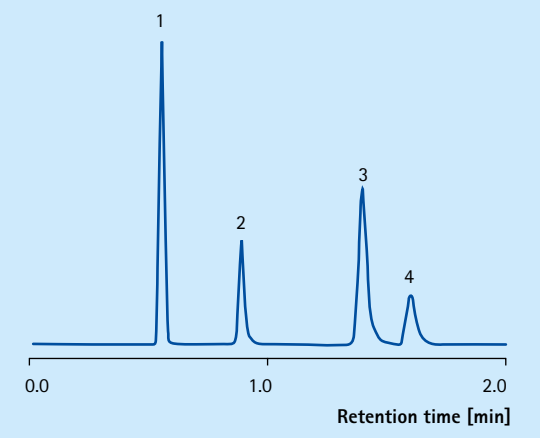
Chromolith® RP-18 endcapped 3 mm i.d. columns: Fast and solvent saving separations at lower flow rates

Chromolith® Performance RP-18 endcapped 100-3 mm is the ideal alternative to conventional particulate columns with internal diameters of 3, 4 or 4.6 mm. Even difficult separations, which often take 15 to 30 minutes on particulate columns, typically only require 5 to 10 minutes on Chromolith® 3 mm. Furthermore, the columns can be easily linked using the column coupler to produce columns of 20 cm or more. As shown below, this results in very high peak resolution at moderate pressure, with flow rates between 1 to 1.5 mL/min.

Chromolith® Performance RP-18e 100-4.6 mm

Column	Chromolith® Performance RP-18 endcapped 100-4.6 mm
Mobile phase	Acetonitrile / water 40/60
Flow rate	4.0 mL/min
Pressure	137 bar
Detection	UV 254 nm 2.4 µL flow cell*
Temperature	ambient
Injection volume	1 µL*
Sample	1. Biphenyl-4,4'-ol 2. Biphenyl-2,2'-ol 3. Biphenyl-4-ol 4. Biphenyl-2-ol

** For optimum results with 3 mm columns, extra-column volume must be small.*

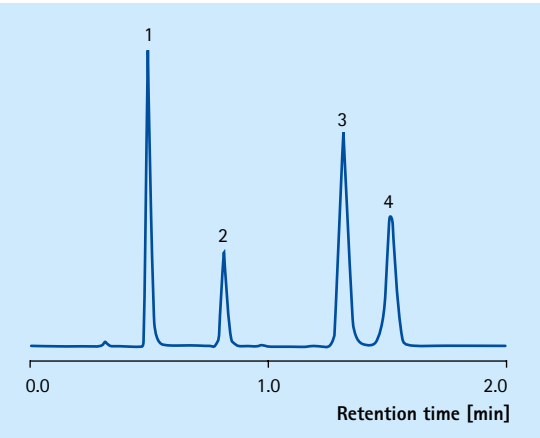


A typical fast separation of four compounds in less than two minutes using a Chromolith® 4.6 mm i.d. column at 4 mL/min.

Chromolith® Performance RP-18e 100-3 mm

Column	Chromolith® Performance RP-18 endcapped 100-3 mm
Mobile phase	Acetonitrile / water 40/60
Flow rate	1.7 mL/min
Pressure	100 bar
Detection	UV 254 nm 2.4 µL flow cell*
Temperature	ambient
Injection volume	1 µL*
Sample	1. Biphenyl-4,4'-ol 2. Biphenyl-2,2'-ol 3. Biphenyl-4-ol 4. Biphenyl-2-ol

** For optimum results with 3 mm columns, extra-column volume must be small.*



The same separation on a Chromolith® 3 mm i.d. column demonstrates improved sensitivity at just 1.7 mL/min. This equates to solvent savings of 57 %.

Chromolith® HighResolution RP-18 endcapped

Chromolith® 4.6 mm i.d. RP-18 endcapped columns: The faster way to trouble-free separations

Chromolith® 4.6 mm i.d. columns represent the most commonly used column dimension. They are compatible with all standard HPLC instruments, and allow a wide range of flow rates, from 0.6 to 4.5 mL/min. These columns are available in two versions: standard columns with 2 µm macropores, and HighResolution columns with 1.15 µm macropores.

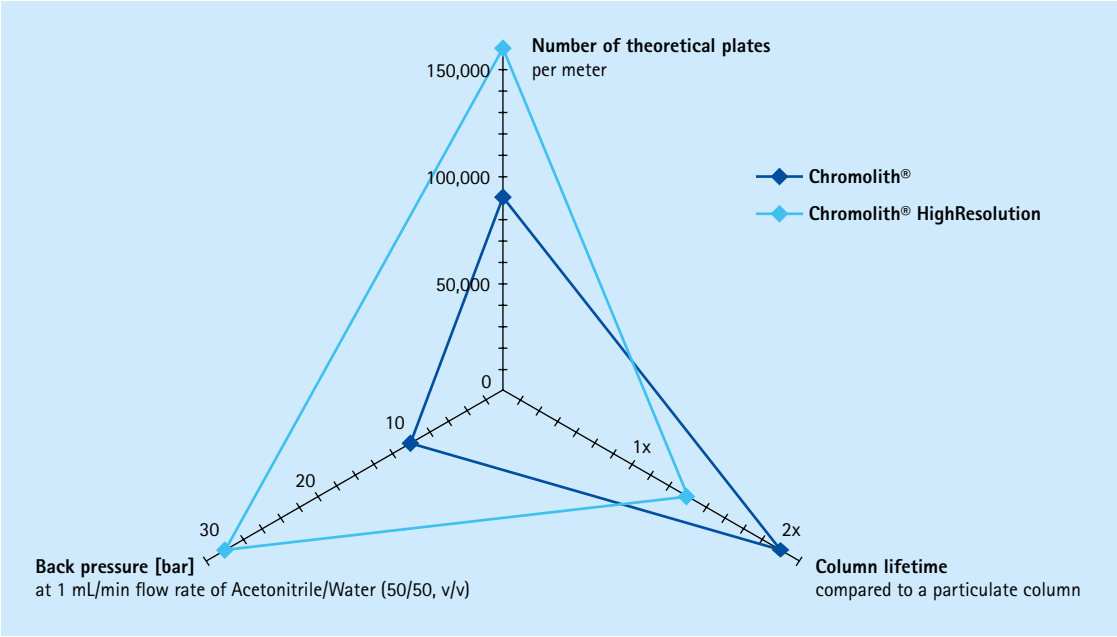
Standard Chromolith® columns have 2 µm macropores, and an efficiency equal to 4.5 µm particulate columns. They allow very high flow rates, extreme throughputs, and the analysis of relatively dirty samples. The lifetime of these columns is particularly long. If necessary, up to ten Chromolith® columns can be coupled in a row to enhance efficiency and resolution.

In contrast, Chromolith® HighResolution (HR) columns possess 1.15 µm macropores, which results in higher efficiency and improved peak shape. Although this causes higher back pressure, it is still less than half that of any particulate column of the similar efficiency.

Chromolith® HighResolution RP-18 endcapped

Comparison: Chromolith® and Chromolith® HighResolution

Chromolith® HighResolution has around 50 % higher efficiency, excellent peak symmetry and still more than 30 % longer lifetime compared with particulate columns. Two Chromolith® HighResolution columns could be easily coupled in order to achieve even higher resolution. The completely endcapped stationary phase enables peak-tailing free elution of basic compounds.

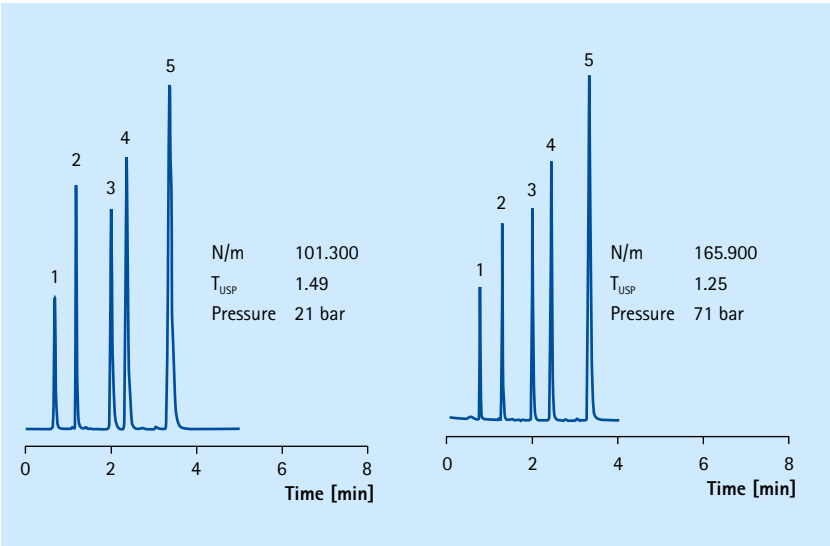


Our classical Chromolith® columns are recommended for analyzing matrix-rich samples, as this type of column will have a longer lifetime. Also, lower back pressure would allow column coupling if necessary.

Higher efficiency, symmetrical peaks

Chromolith® Performance RP-18e 100-4.6 mm

Chromolith® HighResolution RP-18e 100-4.6 mm



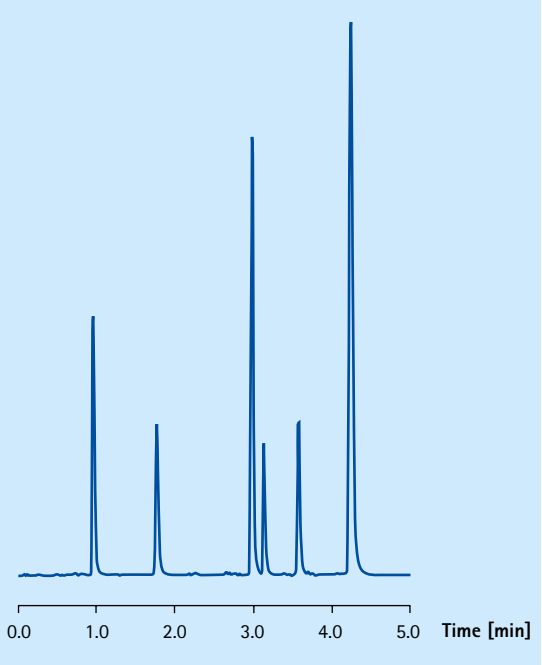
Mobile phase	Acetonitrile / water 60/40
Flow rate	2 mL/min
Detection	UV 254 nm
Temperature	ambient
Injection volume	5 µL
Sample	1. Urea
	2. Biphenyl-2-ol
	3. Progesterone
	4. Hexanophenon
	5. Anthracene

Improved peak shape for basic compounds

The completely endcapped stationary phase enables the elution of basic compounds with significantly less tailing.

Chromolith® HighResolution RP-18e 100-4.6 mm

Column	Chromolith® HighResolution RP-18 endcapped 100-4.6 mm
Mobile phase	A: ACN B: 20 mM NaH_2PO_4 buffer pH 7.6
Gradient	0 min 20 % A 0.5 min 45 % A
Flow rate	2 mL/min
Column pressure	63 – 69 bar
Detection	UV 254 nm
Vol. detector cell	16 µL
Temperature	ambient
Injection volume	1 µL
Sample	1. Caffeine 2. Aniline 3. N-Methylaniline 4. 2-Ethylaniline 5. 4-Nitranisole 6. N,N-Dimethylaniline

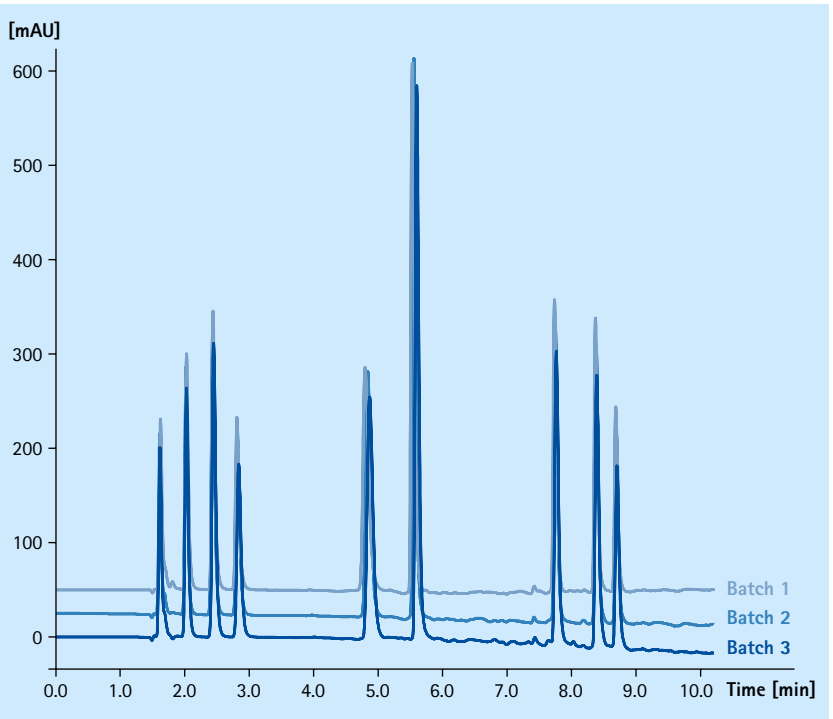


Excellent batch-to-batch reproducibility

The batch-to-batch reproducibility of Chromolith® HPLC columns is strictly controlled and fulfills the requirements of QA and QC laboratories.

Chromolith® HighResolution RP-18e 100-4.6 mm

Column	Chromolith® HighResolution RP-18 endcapped 100-4.6 mm
Mobile phase	A: Acetonitrile + 0.1 % TFA B: Water + 0.1 % TFA
Gradient	2 min 0 % A 10 min 30 % A
Flow rate	1 mL/min
Detection	UV 210 nm
Temperature	25°C
Injection volume	2 µL
Sample	1. Norepinephrine 2. Octopamine 3. Epinephrine tartrate 4. Dopamine 5. DOPA 6. Norephedrine 7. Ephedrine 8. N-Methylephedrine



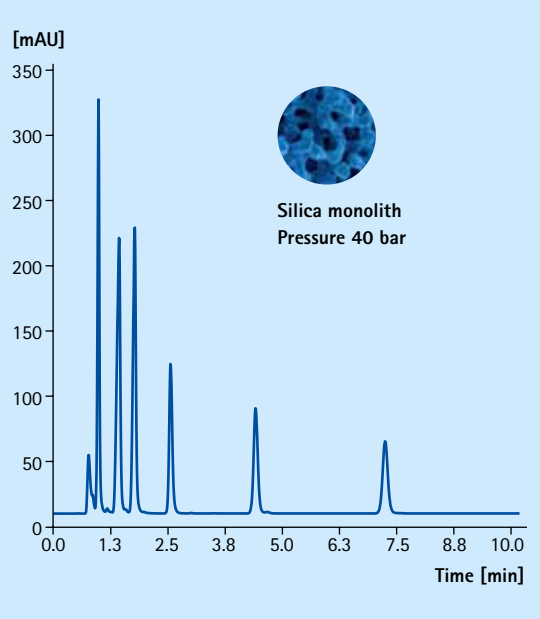
Chromolith® HighResolution RP-18 endcapped

The ideal alternative to sub-3 µm particulate columns

At a flow rate of 1 mL/min, a chromatogram run on a Chromolith® HR column is almost identical to one run on a particulate column with sub-3 µm particles. Chromolith® HR also delivers similar results to a column packed with 2.6 µm i.d. core-shell particles – however at much lower back pressures.

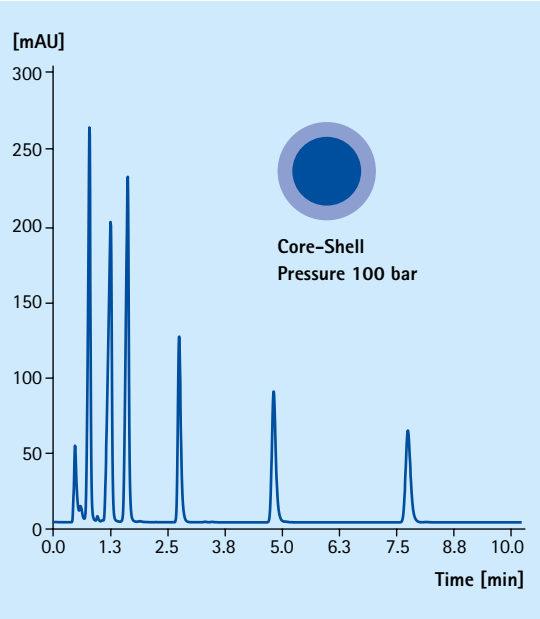
Silica monolith

Column	Chromolith® HighResolution RP-18 endcapped 50-4.6 mm, Silica monolith		
Mobile phase	A: Acetonitrile B: 20 mM Phosphate buffer pH 4.5		
Gradient	Time/min	% A	% B
	0.0	20	80
	12.0	40	60
Flow rate	1.0 mL/min		
Pressure	40 bar		
Detection	UV 230 nm		
Temperature	22°C		
Injection volume	2 µL		
Sample	1. Ascorbic acid 2. 4-Hydroxybenzoic acid 3. Benzoic acid 4. Sorbic acid 5. Methyl 4-hydroxybenzoate 6. Ethyl 4-hydroxybenzoate 7. Propyl 4-hydroxybenzoate		



Core-shell, 2.6 µm particles

Column	Core-shell RP-18 endcapped 50-4.6 mm, 2.6 µm particles		
Mobile phase	A: Acetonitrile B: 20 mM Phosphate buffer pH 4.5		
Gradient	Time/min	% A	% B
	0.0	20	80
	12.0	40	60
Flow rate	1.0 mL/min		
Pressure	100 bar		
Detection	UV 230 nm		
Temperature	22°C		
Injection volume	2 µL		
Sample	1. Ascorbic acid 2. 4-Hydroxybenzoic acid 3. Benzoic acid 4. Sorbic acid 5. Methyl 4-hydroxybenzoate 6. Ethyl 4-hydroxybenzoate 7. Propyl 4-hydroxybenzoate		



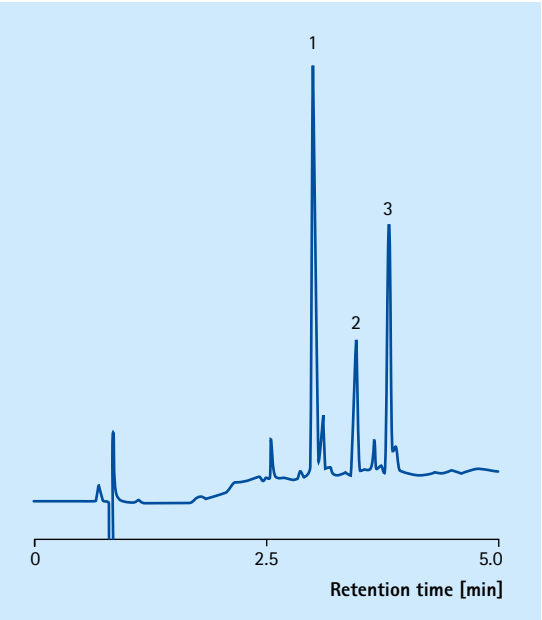
Chromolith® RP-8 endcapped

With its shorter alkyl chain, Chromolith® RP-8 endcapped offers less retention and slightly different selectivity than Chromolith® RP-18 endcapped. Thus, it is possible to achieve a baseline separation on the RP-8 endcapped bonded column, whereas no separation at all is observed under identical elution conditions on a RP-18 endcapped bonded silica column. **Chromolith® RP-8 endcapped HPLC columns offer all the benefits of monolithic silica technology for reversed-phase chromatography.**

Separation examples

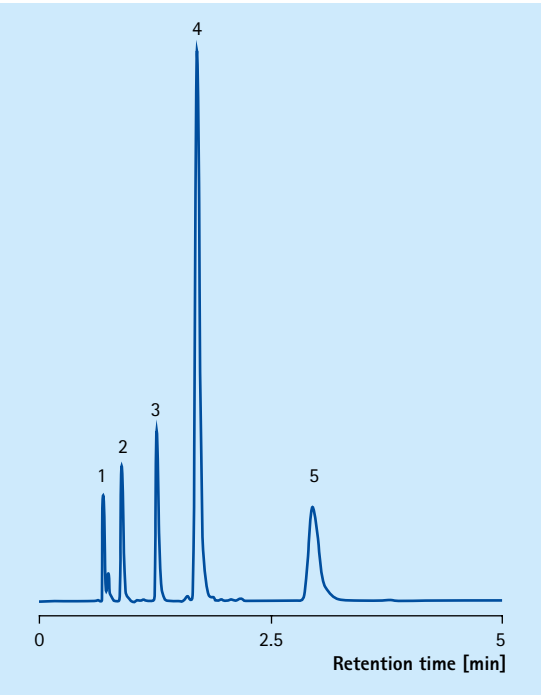
Chromolith® Performance RP-8e 100-4.6 mm

Column	Chromolith® Performance RP-8 endcapped 100-4.6 mm		
Mobile phase	A: Acetonitrile / water 90/10 + 0.1 % TFA B: 0.1 % TFA in water		
Gradient	Time/min	% A	% B
	0.0	45	55
	1.0	90	10
	3.0	90	10
Flow rate	2 mL/min		
Pressure	30 – 40 bar		
Detection	214 nm		
Temperature	ambient		
Injection volume	30 µL		
Sample	1. (Sar1, Ala8)-Angiotensine II		87 µg/mL
	2. (Sar1, Ile8)-Angiotensine II		87 µg/mL
	3. Angiotensine I		47 µg/mL



Chromolith® Performance RP-8e 100-4.6 mm

Column	Chromolith® Performance RP-8 endcapped 100-4.6 mm		
Mobile phase	Methanol / buffer 15/85 v/v (buffer: 1.01 g heptane-sulfonic acid + 24 mL acetic acid (100 %) + 2 mL TEA to 1 L)		
Flow gradient	Time/min	mL/min	
	0	2	
	2	4	
	5	4	
Pressure	40 – 150 bar		
Detection	270 nm		
Temperature	ambient		
Injection volume	10 µL		
Sample	1. Uracil		0.01 mg/mL
	2. Nicotinic acid amid		0.06 mg/mL
	3. Pyridoxine hydrochlorid		0.06 mg/mL
	4. Riboflavine		0.05 mg/mL
	5. Thiamine dichlorid		0.03 mg/mL



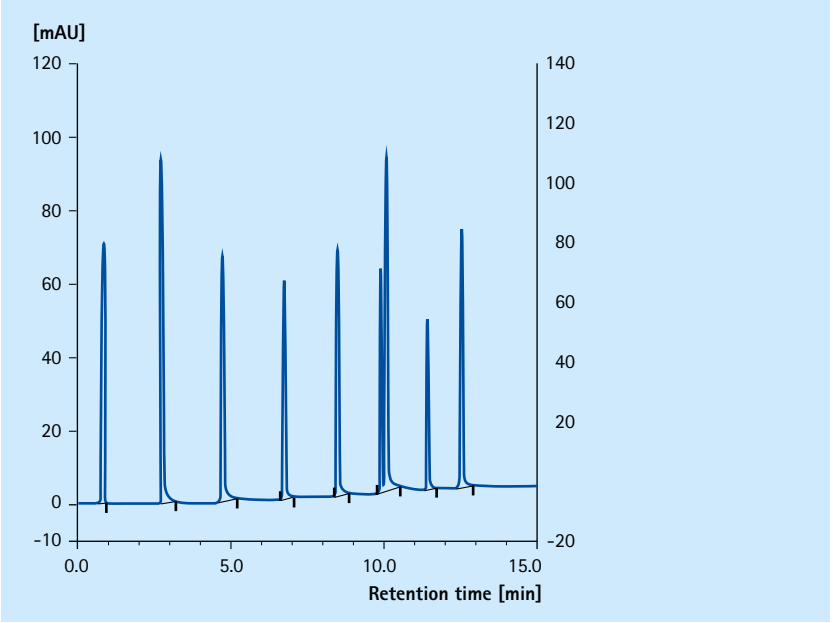
Chromolith® Phenyl

Due to their π-π interactions, Chromolith® Phenyl HPLC columns offer greater selectivity towards aromatic ring-containing compounds than standard alkyl phases. These columns are ideal for the separation of aromatic compounds, flavonoids, fatty acids, PAH, preservatives, purines and pyrimidines.

Separation of alkylphenones using ACN

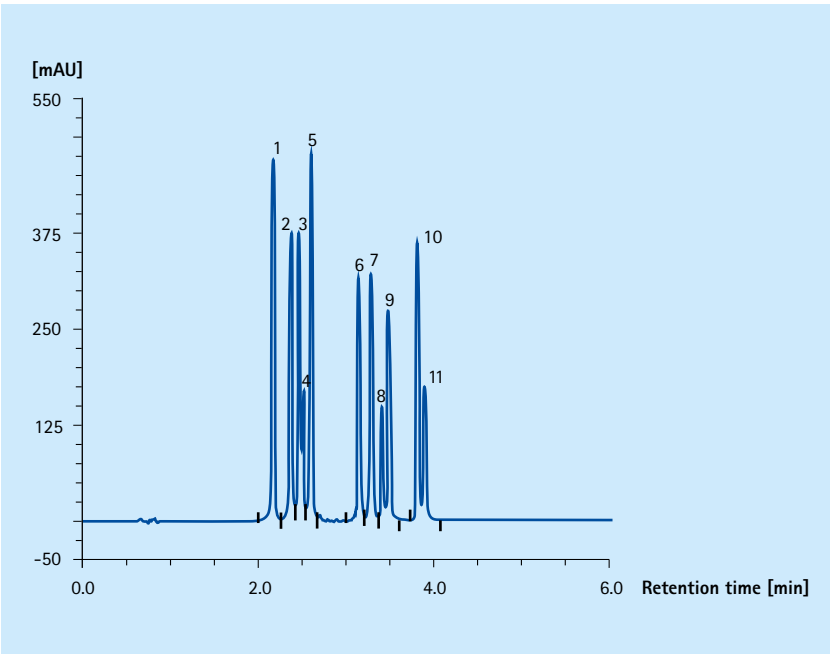
Chromolith® Phenyl 100–4.6 mm

Column	Chromolith® Phenyl 100-4.6 mm
Mobile phase	A: ACN / B: H_2O
Gradient	0 min 22 % A 15 min 85 % A 5 min 95 % A
Flow rate	2 mL/min
Detection	UV 254 nm, response time 0.1 s
Injection volume	2 µL
Sample	1. Thiourea 2. Acetanillide 3. Acetophenone 4. Propionphenone 5. Butyrophenone 6. Benzophenone 7. Valerophenone 8. Hexanophenone 9. Heptanophenone dissolved in 100 mL Methanol/Water 90/10



Chromolith® Phenyl 100–4.6 mm

Column	Chromolith® Phenyl 100-4.6 mm
Mobile phase	A: ACN / B: H_2O
Gradient	0 min 40 % A 60 % B 2.5 min 95 % A 5 % B 5 min 95 % A 5 % B
Flow rate	2 mL/min
Detection	UV 240 nm, response time 0.1 s
Injection volume	1 µL
Sample	1. Fluoxymesterone 2. Boldenone 3. Methandrostenolone 4. Testosterone 5. Methyltestosterone 6. Boldenone acetate 7. Testosterone acetate 8. Nandrolone acetate 9. Testosterone propionate 10. Nandrolone phenylpropionate 11. Testosterone isocaproate in 100 mL ACN/Water

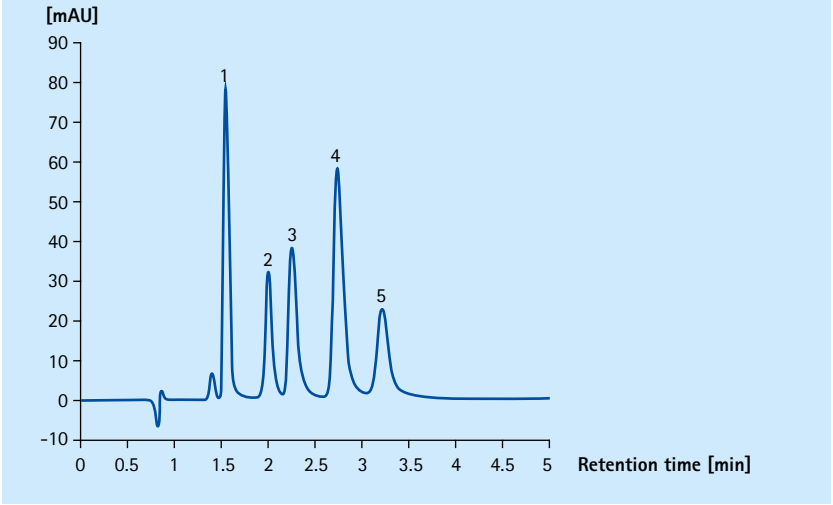


Chromolith® CN

Cyano columns are generally more polar than traditional alkyl silica columns. The functional groups are highly ordered, reducing steric hindrance for the solute. The modification also allows cation exchange activity, which is higher at neutral pH than in acidic conditions. Chromolith® CN columns are suitable for the separation of alkaloids, oils, flavonoids, glycols, fenols, phthalates, steroids and sulfonamides.

Separation example: Five estrogens
Chromolith® CN 100–4.6 mm

Column	Chromolith® CN 100-4.6 mm	
Mobile phase	Methanol/0.1 % TFA 30/70 v/v	
Flow rate	2.0 mL/min	
Detection	220 nm	
Cell volume	11 µL	
Temperature	ambient	
Injection volume	5 µL	
Sample	1. Estriol	9.4 mg/mL
	2. Estradiol	8.7 mg/mL
	3. Testosterone	11.6 mg/mL
	4. Ethynylestradiol	7.9 mg/mL
	5. Estrone	13.3 mg/mL
	solved in 100 mL ACN/$H_2$0 5/5	

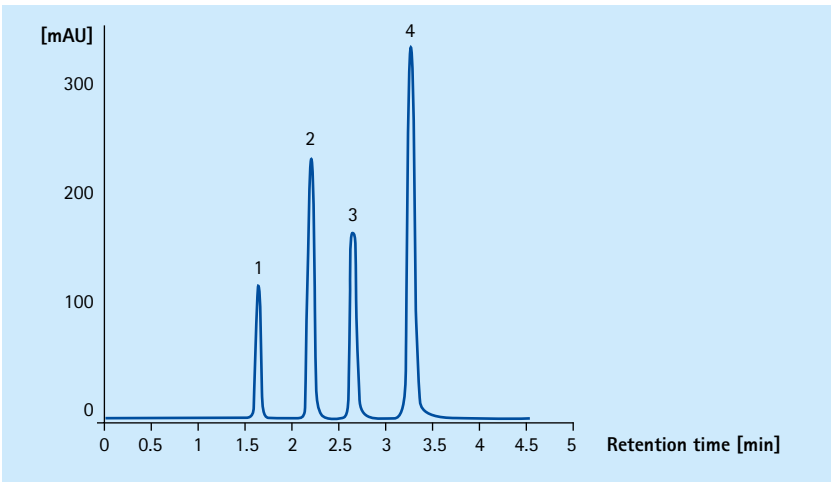


Chromolith® Diol

Chromolith® Diol columns are more versatile than bare silica columns, and often offer improved reproducibility. The bonded phase's hydroxyl groups provide good selectivity without excessive retention. This is due to weaker hydrogen bonding with diol groups than with silanols on a bare silica surface. In aqueous phases, the diol phase can effectively shield the silica surface from interacting with proteins. Diol columns are commonly used for the separation of steroids and sterols under normal-phase conditions. Chromolith® Diol columns are suitable for the separation of alcohols, amino acids, carotinoids, oils, glycols, preservatives, proteins, sugars, sulfonamides, and water-soluble vitamins.

Separation example: Anisole
Chromolith® Diol 100–4.6 mm

Column	Chromolith® Diol 100-4.6 mm	
Mobile phase	n-Heptane / Dioxane 95/5 v/v	
Flow rate	1.3 mL/min	
Detection	254 nm	
Cell volume	11 µL	
Temperature	ambient	
Injection volume	5 µL	
Sample	1. Anisol	390 µg/mL
	2. 3-Nitroanisol	70 µg/mL
	3. 4-Nitroanisol	260 µg/mL
	4. 2-Nitroanisol	180 µg/mL



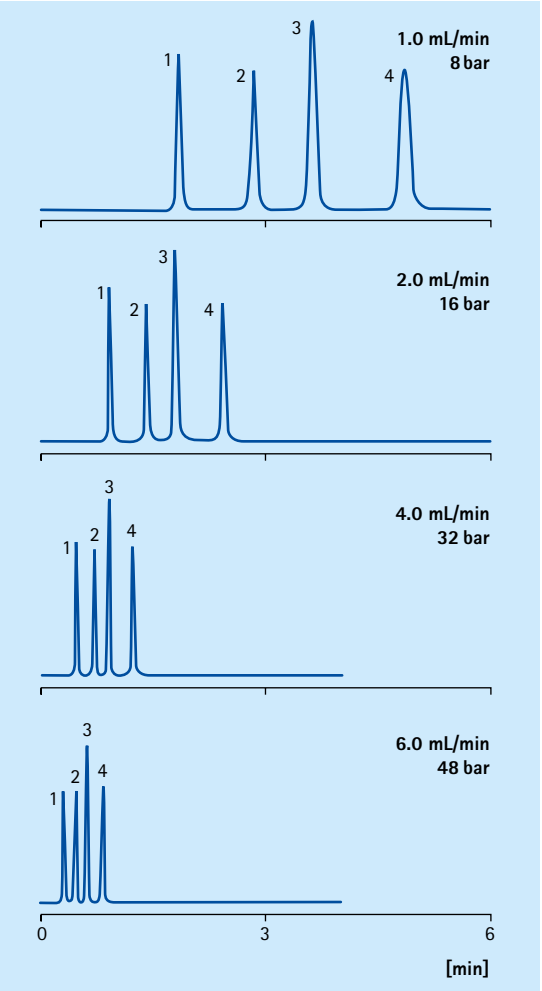
Chromolith® Si

Based on high-purity silica, Chromolith® Si is designed for normal-phase separations of polar non-ionic organic compounds. The column offers all of the benefits of monolithic silica technology.

Separation examples

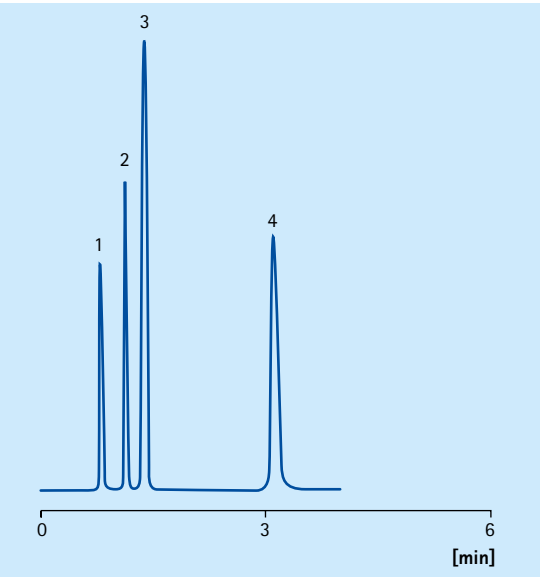
Chromolith® Performance Si 100–4.6 mm

Column	Chromolith® Performance Si 100-4.6 mm	
Mobile phase	n-Heptane / Dioxane 95/5 v/v	
Flow rate	2 mL/min	
Pressure	14 bar	
Detection	254 nm	
Temperature	ambient	
Injection volume	5 µL	
Sample	1. Anisole	0.39 mg/mL
	2. 3-Nitroanisole	0.07 mg/mL
	3. 4-Nitroanisole	0.26 mg/mL
	4. 2-Nitroanisole	0.18 mg/mL



Chromolith® Performance Si 100–4.6 mm

Column	Chromolith® Performance Si 100-4.6 mm	
Mobile phase	n-Heptane / Dioxane 95/5 v/v	
Flow rate	2 mL/min	
Pressure	14 bar	
Detection	254 nm	
Temperature	ambient	
Injection volume	10 µL	
Sample	1. Toluene	0.16 mg/mL
	2. Nitrobenzene	0.02 mg/mL
	3. 2,3-Dimethylanthraquinone	0.02 mg/mL
	4. 2-Nitroacetanilide	0.10 mg/mL



Chromolith® NH_2

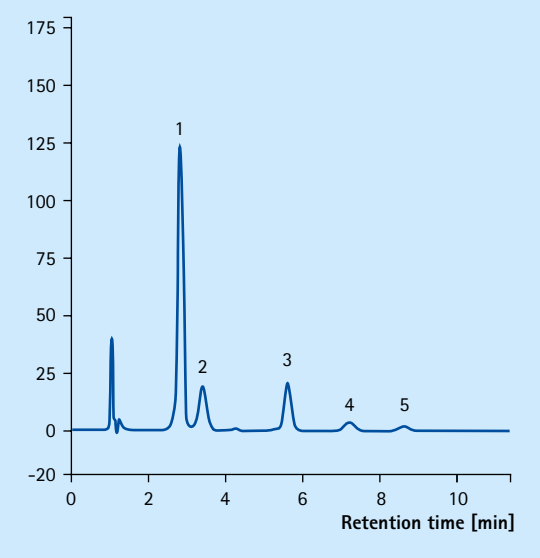
Chromolith® aminopropyl-modified columns possess medium polarity, between those of bare (normal-phase) silica and reversed-phase silica. Consequently, they display hydrophilic as well as hydrophobic properties, and can be used under both reversed-phase and normal-phase conditions. However, retention is weaker than on silica or RP supports. In acidic solutions, the NH_2 groups are protonated ($-NH_3^+X^-$) and display the characteristics of a weak anion exchanger. Hence, the columns can also be used as ion exchangers.

Chromolith® NH_2 columns offer high matrix tolerance and analysis speed, as well as an extended lifetime within the pH range of 2.5 to 7.5. These columns are suitable for the separation of anions, organic acids, and carbohydrates (mono and disaccharides, such as fructose, glucose, sucrose, maltose and lactose).

Separation example

Chromolith® Performance NH_2 100–4.6 mm

Column	Chromolith® Performance NH_2 100-4.6 mm	
Mobile phase	Acetonitrile / Water 80/20	
Flow rate	1.5 mL/min	
Pressure	9 bar	
Detection	190 nm	
Detector cell volume	16 µL	
Temperature	23°C	
Injection volume	10 µL	
Sample	1. Fructose	53.71 mg
	2. Glucose	46.38 mg
	3. Sucrose	68.75 mg
	4. Maltose	15.71 mg
	5. Lactose	62.05 mg



Preparative columns
10 mm / 25 mm i.d.

Offering faster sample throughput at lower pressure, Chromolith® Prep and SemiPrep HPLC columns are ideal for direct scale-up from analytical to prep and semi-prep. The excellent accessibility of the mesopores (total porosity > 80 %), and the short diffusion length inside the pores ensure fast adsorption and desorption kinetics. This leads to faster separations and higher productivity. The monolithic structure of Chromolith® SemiPrep and Prep columns also eliminates inlet bed settling or bed splitting under high pressure. Column reliability, reproducibility and extended lifetime are ensured.

Benefits of preparative columns

- Direct scale-up from analytical to semi-prep or prep columns
- Faster sample throughput at lower operating pressure compared to semi/prep columns packed with 5 µm particles
- Sharp separations, even at high sample loading
- Excellent column lifetime due to rugged monolithic silica structure
- Higher porosity allows fast adsorption and desorption kinetics
- Compared to particulate sorbents, monolithic columns ensure shorter separation times and less solvent consumption
- Higher productivity and greater efficiency than particulate sorbents

Chapter content

Chromolith® SemiPrep

Perfect scale-up from analytical to preparative LC

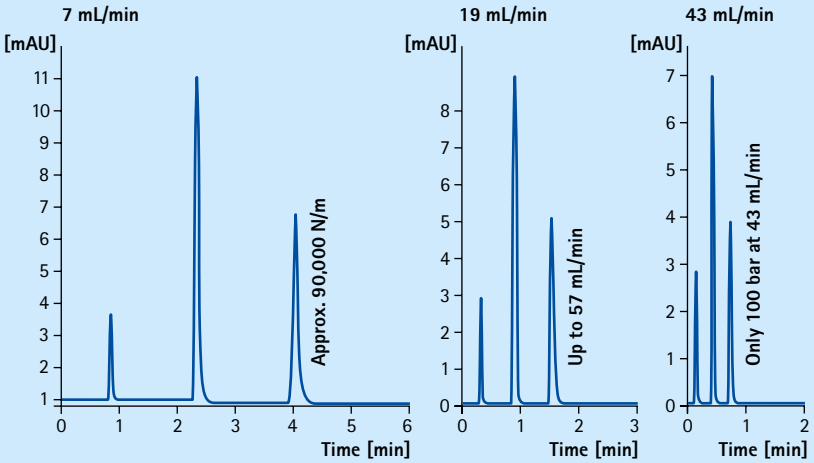
Ready-to-use Chromolith® SemiPrep column.

Optimum separation at flow rates exceeding 40 mL/min

Chromolith® SemiPrep HPLC columns are ideally suited for direct scale-up from analytical to semi-prep. This is because they offer faster sample throughput at a lower operating pressure compared to semi-prep columns packed with 5 µm particles.

Chromolith® SemiPrep 10 mm i.d. columns combine high separation speed with excellent performance. This makes them the perfect alternative to particulate columns of 10 mm i.d. (and even 21.2 mm i.d.). They have the same bimodal porous silica rod structure as Chromolith® analytical columns with an internal diameter of 4.6 mm. Their macropores are 2 µm in diameter and the mesopores are 13 nm. This combination dramatically reduces separation time while increasing efficiency.

Acetonitrile/water 60/40, Data for anthracene (3rd peak)



Separation of a standard mixture

Chromolith® SemiPrep C18e, 100–10 mm

Mobile phase	Acetonitrile / water 60/40
Flow rate	2 mL/min
Detection	UV 254 nm
Temperature	ambient
Injection volume	5 µL
Sample	1. Thiourea
	2. Progesterone
	3. Anthracene

Chromolith® Prep
Higher speed, efficiency and productivity

Preparative HPLC involves much higher sample volumes than analytical chromatography. Consequently, greater sample throughput and separation speed are essential for optimal productivity. These criteria are best fulfilled by Chromolith® Prep columns. The combination of macro and mesopores maximizes separation efficiency and flow rate, while minimizing resistance.

The formula for direct scale-up

Analytical separations can be easily transferred to Chromolith® SemiPrep and Prep columns by linear transfer of methods. The objective of any preparative separation strategy is high sample throughput per unit of time. Therefore, columns are often run under concentration and/or volume overload conditions. However, the maximum load on the column is dependent on the complexity of the separation and the nature of the sample. Whether working in a linear or non-linear mode, the flow rate or injection volume is calculated acc. to the equation below.

$$\frac{X_{an}}{\pi r_{an}^2} = \frac{X_{pr}}{\pi r_{pr}^2} \cdot \frac{1}{c_L}$$

X_{an}	Flow rate in the analytical system	
X_{pr}	Flow rate in the preparative system	$X_{pr} = X_{an} \cdot r^2_{pr} \cdot c_L / r^2_{an}$
r_{an}	Radius of analytical column	
r_{pr}	Radius of preparative column	
c_L	Length of the preparative column to length of the analytical column	
M	Substance mass	$M_{pr} = M_{an} \cdot r^2_{pr} \cdot c_L / r^2_{an}$

Guide values of typical flow rates and loading capacity for transfer from an analytical to a preparative column

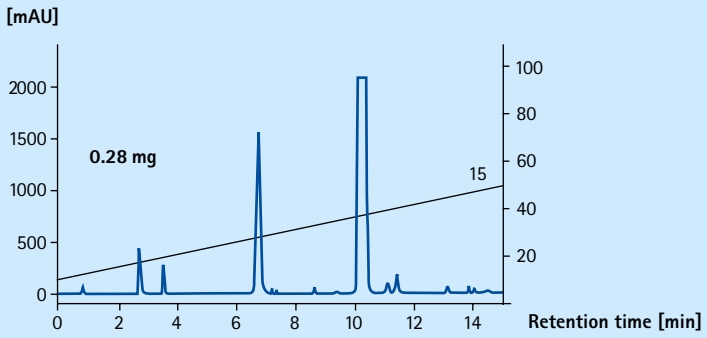
Columns	Column dimension [length / diameter]	Typical flow rate	Loading capacity	Loading volume
Analytical column	100 – 4.6 mm	2 mL/min	5 mg	5 – 50 µL
Preparative column	100 – 25 mm	60 mL/min	150 – 370 mg	100 – 1,500 µL

Ready-to-use Chromolith® Prep column.

Analytical separation

Chromolith® Performance RP-18e 100-4.6 mm

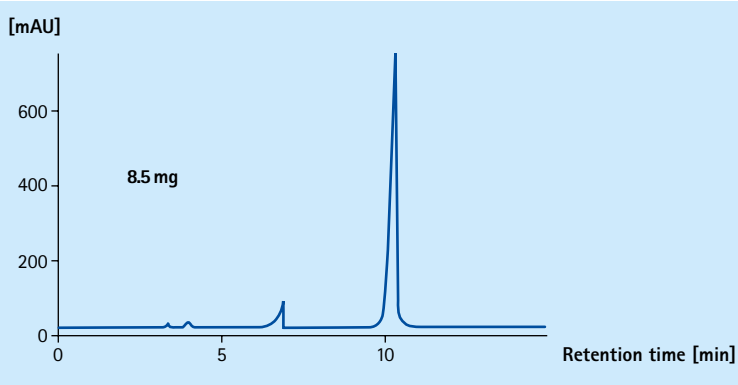
Column	Chromolith® Performance RP-18 endcapped 100-4.6 mm
Mobile phase	A: Water + 0.1 % formic acid B: Acetonitrile
Gradient	linear gradient from 10 % B to 40 % in 14 min
Flow rate	2 mL/min
Detection	UV 254 nm
Sample	**0.28 mg** Heterocyclic racemate (EMD 53986) in 10 µL DMSO



Preparative separation

Chromolith® Prep RP-18e 100-25 mm

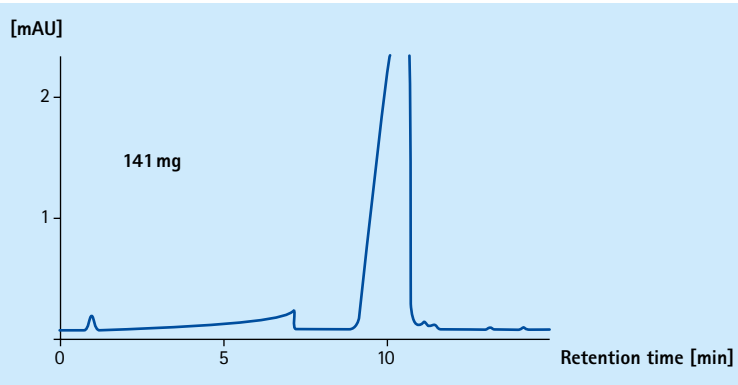
Column	Chromolith® Prep RP-18 endcapped 100-25 mm
Mobile phase	A: Water + 0.1 % formic acid B: Acetonitrile
Gradient	linear gradient from 10 % B to 40 % in 14 min
Flow rate	60 mL/min
Detection	UV 254 nm
Sample	**8.46 mg** Heterocyclic racemate (EMD 53986) in 300 µL DMSO



Preparative separation

Chromolith® Prep RP-18e 100-25 mm

Column	Chromolith® Prep RP-18 endcapped 100-25 mm
Mobile phase	A: Water + 0.1 % formic acid B: Acetonitrile
Gradient	linear gradient from 10 % B to 40 % in 14 min
Flow rate	60 mL/min
Detection	UV 254 nm
Sample	**141 mg** Heterocyclic racemate (EMD 53986) in 300 µL DMSO



Various applications with Chromolith® Prep monolithic columns

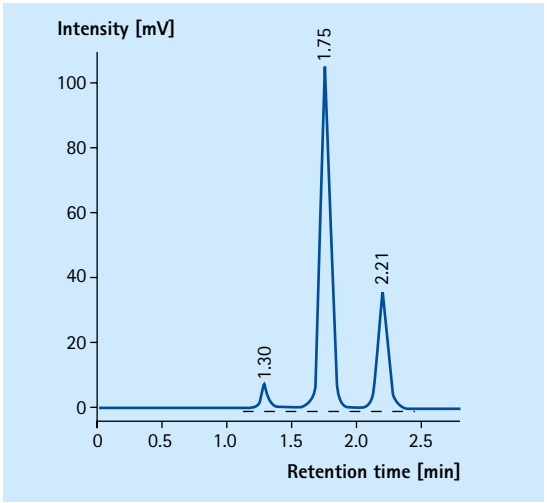
Comparison of flow rates

Chromolith® Prep columns can be operated at a flow rate of up to 400 mL/min, and pressures of up to 100 bar. This is a tenfold increase in flow rate compared to particulate columns of an equivalent size.

Separation at different flow rates 40 and 390 mL/min

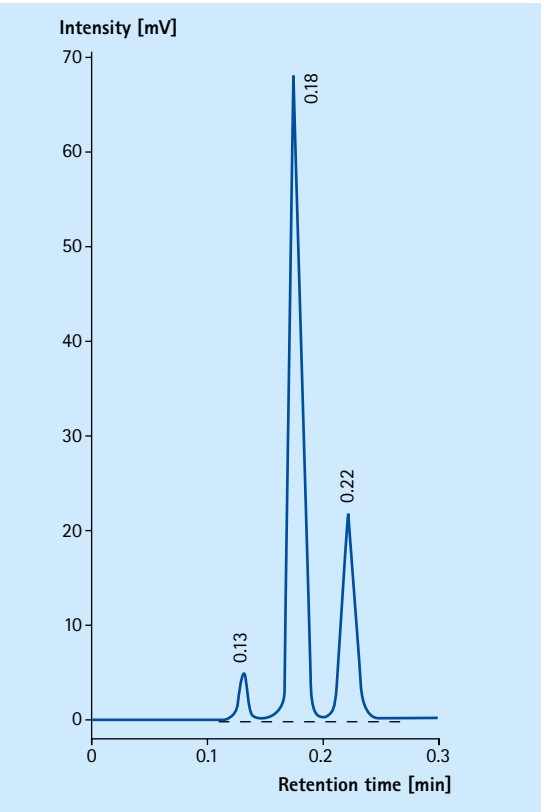
Chromolith® Prep Si 100–25 mm

Column	Chromolith® Prep Si 100-25 mm
Solvent	n-Heptane / Dioxane (80/20 v/v)
Flow rate	40 mL/min
Sample	1. Toluene
	2. Dimethylphthalate
	3. Dibutylphthalate



Chromolith® Prep Si 100–25 mm

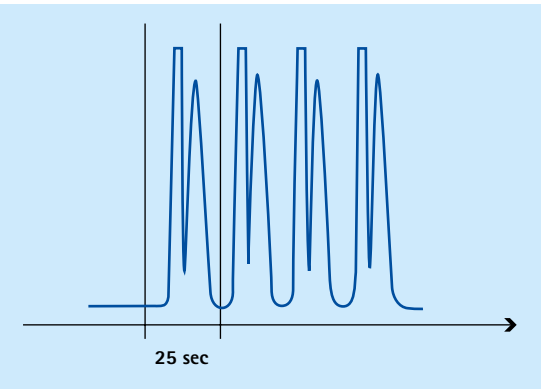
Column	Chromolith® Prep Si 100-25 mm
Solvent	n-Heptane / Dioxane (80/20 v/v)
Flow rate	390 mL/min
Sample	1. Toluene
	2. Dimethylphthalate
	3. Dibutylphthalate



Separation of diastereomers with a productivity of 861 g/d

Chromolith® Prep Si 100–25 mm

Column	Chromolith® Prep Si 100-25 mm
Solvent	n-Heptane / Dioxane (80/20 v/v)
Flow rate	140 mL/min
Injection	249 mg
Cycle time	25 sec
Sample	Fluoro-dihydro-oxyranyl-benzopyran



Chromolith®
HPLC guard cartridges and kits

Although monolithic columns are well known for their robustness and longevity, Merck Millipore's Chromolith® guard cartridges and kits further enhance these advantages. The guard columns are chemically modified with hydrophobic n-octadecyl (C18) groups on the surface of the monolithic silica rod, making them suitable for reversed-phase chromatography.

Guard cartridges

Chromolith® HPLC guard cartridges are extremely easy to use. They are simply added directly in front of the main column to protect it from chemical or mechanical contamination. Due to the benefits of monolithic technology, and the convenience of Chromolith® guard columns, they are also popular for use with classical particulate columns. Moreover, guard columns can be used as trap columns when large sample volumes are to be injected. Guard columns should be changed frequently in order to avoid excessive accumulation of impurities.

Guard cartridge starter kit

The Chromolith® guard cartridge kit includes everything needed to significantly enhance the lifetime of monolithic columns: a guard cartridge holder, and three guard cartridges.

a) b) c) d) e)

For maximum convenience and flexibility, guard cartridges are available in five dimensions with corresponding holders made of PEEK or stainless steel, having different maximum back pressures.

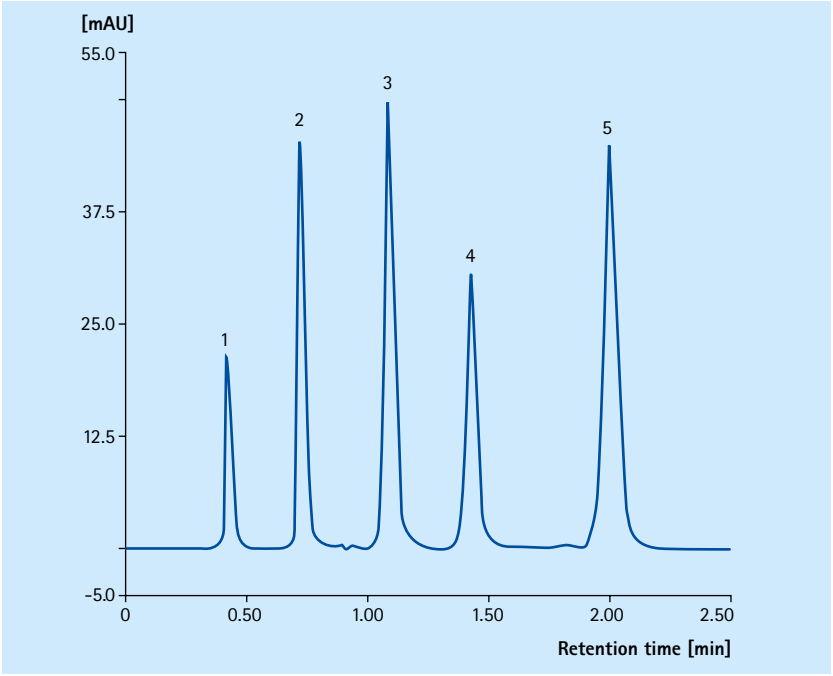
Guard cartridge holder type	Material holder is made of	Max. back pressure	How to tighten holder	Guard cartridge i.d.	Guard cartridge length
a)	PEEK	200 bar (2,940 psi)	Finger-tight	2 mm,	5 mm
b)	Alumina / PEEK	200 bar (2,940 psi)	Finger-tight + tool (included)	4.6 mm	5 mm, 10 mm
c)	SS	400 bar (5,880 psi)	Finger-tight + tool (not included)	4.6 mm	5 mm, 10 mm
d)	PEEK / SS	150 bar (2,205 psi)	Finger-tight + tool (not included)	10 mm	10 mm
e)	PEEK	100 bar (1,470 psi)	Finger-tight + tool (included)	25 mm	10 mm

PEEK = Poly Ether Ether Ketone, SS = Stainless Steel

Separation examples with and without a pre-column

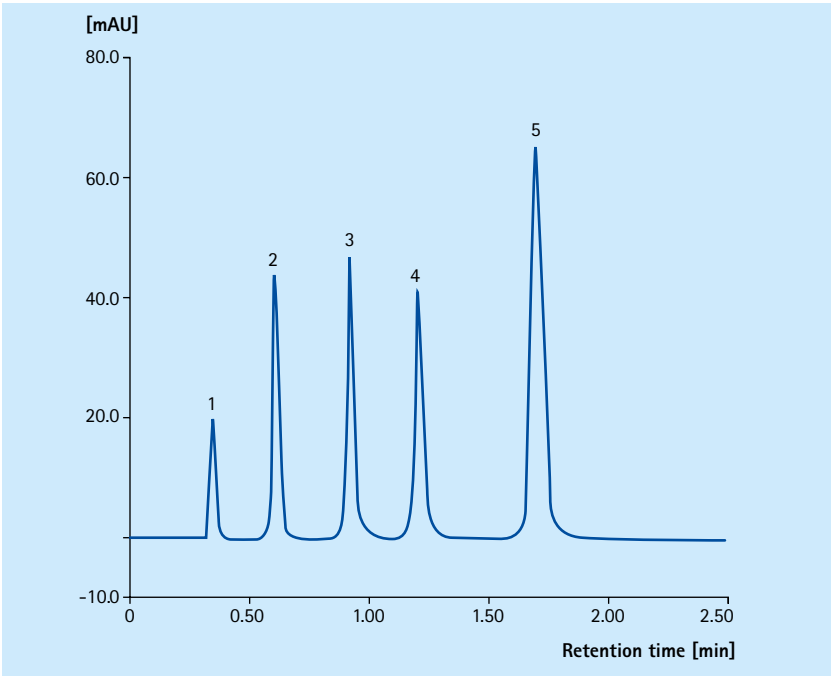
Chromolith® Performance RP-18e 100-2 mm with a Chromolith® RP-18e 5-2 mm pre-column

Column	Chromolith® Performance RP-18e 100-2 mm with pre-column Chromolith® RP-18e 5-2 mm	
Mobile phase	Acetonitrile / water 60/40	
Flow rate	0.38 mL/min	
Pressure	20 bar	
Detection	UV 254 nm	
Anthracene	N/m	113540
	T_{USP}	1.14
	K´-value	3.79
Sample	1. Thiourea	
	2. Biphenyl-2-ol	
	3. Progesterone	
	4. Hexanophenone	
	5. Anthracene	



Chromolith® Performance RP-18e 100-2 mm without a pre-column

Column	Chromolith® Performance RP-18e 100-2 mm without pre-column	
Mobile phase	Acetonitrile / water 60/40	
Flow rate	0.38 mL/min	
Pressure	20 bar	
Detection	UV 254 nm	
Anthracene	N/m	115460
	T_{USP}	1.07
	K´-value	3.90
Sample	1. Thiourea	
	2. Biphenyl-2-ol	
	3. Progesterone	
	4. Hexanophenone	
	5. Anthracene	



As seen in the examples, guard columns have very little negative effects on the separation.
These may include a slight shift in elution time, or a minimal loss in efficiency.

Chromolith®
Column coupler

The Chromolith® HPLC column coupler is designed for linking several monolithic columns together in order to further increase separation efficiency and column performance. The combination results in a theoretical plate count that is significantly higher than any particulate column available. At the same time, pressure is kept well below the HPLC system limit.

The superior column performance achieved by using the Chromolith® column coupler allows you to solve highly critical separation problems in which resolution is a limiting factor. This makes column coupling perfect for chromatographic separations of typically non-separable, complex mixtures.

Typical column efficiency using the Chromolith® column coupler

Column	Length [mm]	Pressure * [bar]	Plate number per column [Anthracene]
Chromolith® Performance 1x	100	30	10,000
Chromolith® Performance 2x	200	60	19,000
Chromolith® Performance 3x	300	90	27,000
Chromolith® Performance 4x	400	120	35,000
Chromolith® Performance 5x	500	150	41,000
Particulate column (5 µm)	250	220	18,500
Particulate column (3.5 µm)	150	400	19,000

Pressure * = 3 mL/min 75 % acetonitrile, 25 % water

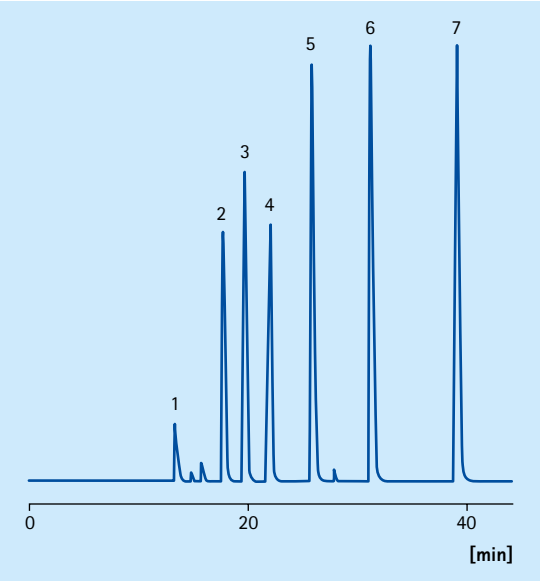
The table shows a comparison between Chromolith® HPLC columns and particulate columns.
The coupling of just two Chromolith® Performance RP-18 endcapped columns yields a separation efficiency of 19,000 theoretical plates per column, which is usually the maximum for particulate columns.

Column coupler

Application of Chromolith® column coupler

81,000 plates at 85 bar pressure

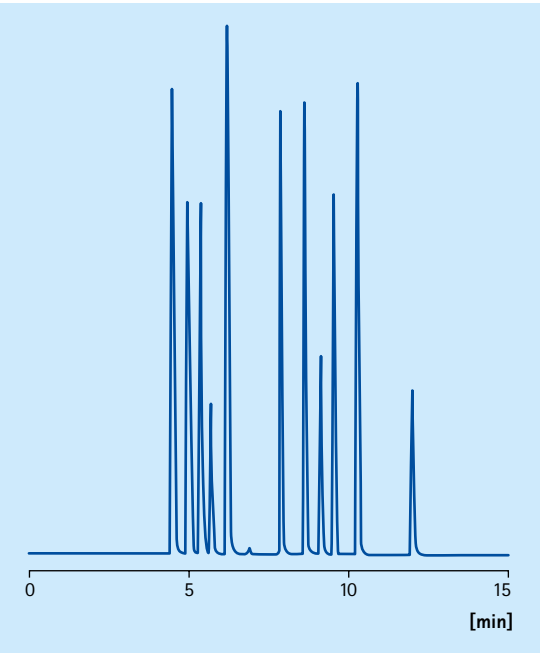
Column	10 columns of Chromolith® Performance RP-18 endcapped 100-4.6 mm
Mobile phase	80 /20 Acetonitrile / water
Flow rate	1 mL/min
Detection	UV 254 nm
Temperature	ambient
Injection volume	10 µL
Sample	1. Thiourea 2. Benzene 3. Toluene 4. Ethylbenzene 5. Propylbenzene 6. Butylbenzene 7. Penylbenzene



Column coupling: 11 steroids

Chromolith® HighResolution RP-18e
2 x 100-4.6 mm / 1 x 50-4.6 mm

Column	Chromolith® HighResolution RP-18 endcapped 2 x 100-4.6 mm / 1 x 50-4.6 mm		
Mobile phase	ACN / water		
Gradient	t [min]	ACN [%]	Water [%]
	0	55	45
	2	95	5
	15	95	5
Flow rate	1 mL/ min		
Column pressure	30 – 68 bar		
LC system	LaChrom® L7000		
Detection	UV = 240 nm		
Vol. detector cell	16 µL		
Temperature	ambient		
Injection volume	10 µL		
Sample	1. Fluoxymesterone 2. Boldenone 3. Methandrostenolone 4. Testosterone 5. Methyltestosterone 6. Boldenone-Acetate 7. Testosterone-Acetate 8. Nandrolone-Propionate 9. Testosterone-Propionate 10. Nandrolone-Phenylpropionate 11. Testosterone-Isocaproate		



Ordering information

Capillary columns

Product	Modification	I.d.	Length	Type	Content	Ord. No.
Chromolith® CapRod®	RP-18e	0.05 mm	150 mm		1 analytical column	1.50403.0001
Chromolith® CapRod®	RP-8e	0.1 mm	150 mm		1 analytical column	1.50400.0001
Chromolith® CapRod®	RP-18e Trap	0.1 mm	50 mm		1 trapping column	1.50426.0001
Chromolith® CapRod®	RP-18e	0.1 mm	150 mm		1 analytical column	1.50402.0001
Chromolith® CapRod®	RP-18e	0.1 mm	300 mm		1 analytical column	1.50424.0001
Chromolith® CapRod® HR	RP-18e	0.1 mm	150 mm		1 analytical column	1.50404.0001
Chromolith® CapRod®	RP-18e Trap	0.2 mm	50 mm		1 trapping column	1.50409.0001
Chromolith® CapRod®	RP-18e	0.2 mm	150 mm		1 analytical column	1.50405.0001
Chromolith® CapRod® HR	RP-18e	0.2 mm	150 mm		1 analytical column	1.50407.0001

Analytical columns

Product	Modification	I.d.	Length	Type	Content	Ord. No.
Chromolith® Performance	RP-18e	2 mm	100 mm		1 HPLC column	1.52006.0001
Chromolith® FastGradient	RP-18e	2 mm	50 mm		1 HPLC column	1.52007.0001
Chromolith® Flash	RP-18e	2 mm	25 mm		1 HPLC column	1.52014.0001
Chromolith® Guard Cartridge Kit	RP-18e	2 mm	5 mm	a*	3 guard cartridges, 1 cartridge holder	1.52008.0001
Chromolith® Guard Cartridge	RP-18e	2 mm	5 mm	a*	3 guard cartridges	1.52009.0001
Chromolith® Performance	RP-18e	3 mm	100 mm		1 HPLC column	1.52001.0001
Chromolith® FastGradient	RP-18e	3 mm	50 mm		1 HPLC column	1.52002.0001
Chromolith® Flash	RP-18e	3 mm	25 mm		1 HPLC column	1.52003.0001
Chromolith® Guard Cartridge Kit	RP-18e	3 mm	5 mm	a*	3 guard cartridges, 1 cartridge holder	1.52004.0001
Chromolith® Guard Cartridge	RP-18e	3 mm	5 mm	a*	3 guard cartridges	1.52005.0001
Chromolith® Performance	RP-18e	4.6 mm	100 mm		1 HPLC column	1.02129.0001
Chromolith® SpeedROD	RP-18e	4.6 mm	50 mm		1 HPLC column	1.51450.0001
Chromolith® Flash	RP-18e	4.6 mm	25 mm		1 HPLC column	1.51463.0001
Chromolith® Guard Cartridge Kit	RP-18e	4.6 mm	10 mm	b*	3 guard cartridges, 1 cartridge holder	1.51471.0001
Chromolith® Guard Cartridge	RP-18e	4.6 mm	10 mm	b/c*	3 guard cartridges	1.51452.0001
Chromolith® Guard Cartridge Kit	RP-18e	4.6 mm	5 mm	b*	3 guard cartridges, 1 cartridge holder	1.51470.0001
Chromolith® Guard Cartridge	RP-18e	4.6 mm	5 mm	b/c*	3 guard cartridges	1.51451.0001
Chromolith® HR	RP-18e	4.6 mm	25 mm		1 HPLC column	1.52020.0001
Chromolith® HR	RP-18e	4.6 mm	50 mm		1 HPLC column	1.52021.0001
Chromolith® HR	RP-18e	4.6 mm	100 mm		1 HPLC column	1.52022.0001
Chromolith® HR Guard Cartridge Kit	RP-18e	4.6 mm	5 mm	b*	3 guard cartridges, 1 cartridge holder	1.52024.0001
Chromolith® HR Guard Cartridge	RP-18e	4.6 mm	5 mm	b/c*	3 guard cartridges	1.52025.0001
Chromolith® Validation Kit	RP-18e	4.6 mm	100 mm		3 columns from 3 different batches	1.51466.0001
Chromolith® Performance	RP-8e	4.6 mm	100 mm		1 HPLC column	1.51468.0001
Chromolith® Guard Cartridge Kit	RP-8e	4.6 mm	5 mm	b*	3 guard cartridges, 1 cartridge holder	1.52012.0001
Chromolith® Guard Cartridge	RP-8e	4.6 mm	5 mm	b/c*	3 guard cartridges	1.52013.0001

* Guard column type examples and detailed information please find on page 34.

Product	Modification	I.d.	Length	Type	Content	Ord. No.
Chromolith®	Phenyl	4.6 mm	25 mm		1 HPLC column	1.52056.0001
Chromolith®	Phenyl	4.6 mm	50 mm		1 HPLC column	1.52057.0001
Chromolith®	Phenyl	4.6 mm	100 mm		1 HPLC column	1.52058.0001
Chromolith® Guard Cartridge	Phenyl	4.6 mm	5 mm		3 guard cartridges	1.52059.0001
Chromolith®	CN	4.6 mm	25 mm		1 HPLC column	1.52046.0001
Chromolith®	CN	4.6 mm	50 mm		1 HPLC column	1.52047.0001
Chromolith®	CN	4.6 mm	100 mm		1 HPLC column	1.52048.0001
Chromolith® Guard Cartridge	CN	4.6 mm	5 mm	b/c*	3 guard cartridges	1.52050.0001
Chromolith®	DIOL	4.6 mm	25 mm		1 HPLC column	1.53170.0001
Chromolith®	DIOL	4.6 mm	50 mm		1 HPLC column	1.53171.0001
Chromolith®	DIOL	4.6 mm	100 mm		1 HPLC column	1.53172.0001
Chromolith® Guard Cartridge	DIOL	4.6 mm	5 mm	b/c*	3 guard cartridges	1.53175.0001
Chromolith® Performance	Si	4.6 mm	100 mm		1 HPLC column	1.51465.0001
Chromolith® Guard Cartridge Kit	Si	4.6 mm	5 mm	b*	3 guard cartridges, 1 cartridge holder	1.52010.0001
Chromolith® Guard Cartridge	Si	4.6 mm	5 mm	b/c*	3 guard cartridges	1.52011.0001
Chromolith® Performance	NH_2	4.6 mm	100 mm		1 HPLC column	1.52028.0001
Chromolith® SpeedROD	NH_2	4.6 mm	50 mm		1 HPLC column	1.52027.0001
Chromolith® Flash	NH_2	4.6 mm	25 mm		1 HPLC column	1.52026.0001
Chromolith® Guard Cartridge Kit	NH_2	4.6 mm	5 mm	b*	3 guard cartridges, 1 cartridge holder	1.52029.0001
Chromolith® Guard Cartridge	NH_2	4.6 mm	5 mm	b/c*	3 guard cartridges	1.52030.0001
Chromolith® Guard Cartridge Holder	-	4.6 mm	5 mm	c*	1 cartridge holder	1.52032.0001
Chromolith® Guard Cartridge Holder	-	4.6 mm	10 mm	c*	1 cartridge holder	1.52033.0001

Semi-preparative and preparative columns

Product	Modification	I.d.	Length	Type	Content	Ord. No.
Chromolith® SemiPrep	RP-18e	10 mm	100 mm		1 HPLC column	1.52016.0001
Chromolith® SemiPrep Guard Cartridge	RP-18e	10 mm	10 mm	d*	3 guard cartridges	1.52036.0001
Chromolith® SemiPrep	Si	10 mm	100 mm		1 HPLC column	1.52015.0001
Chromolith® SemiPrep Guard Cartridge	Si	10 mm	10 mm	d*	3 guard cartridges	1.52035.0001
Chromolith® SemiPrep Guard Cartridge holder	-	10 mm	10 mm	d*	1 cartridge holder	1.52037.0001
Chromolith® Column Coupler	-	-	-		1 column coupler	1.51467.0001
Chromolith® Prep	Si	25 mm	100 mm		1 HPLC column, 2 connectors (1/8" and 1/16")	1.25251.0001
Chromolith® Prep	RP-18e	25 mm	100 mm		1 HPLC column, 2 connectors (1/8" and 1/16")	1.25252.0001
Chromolith® Prep guard cartridge	Si	25 mm	10 mm	e*	1 guard cartridge	1.25260.0001
Chromolith® Prep guard cartridge	RP-18e	25 mm	10 mm	e*	1 guard cartridge	1.25261.0001
Chromolith® Prep sealing set		25 mm			2 O-rings	1.25254.0001
Chromolith® Prep tool set		25 mm			1 mounting tool filter, 1 mounting tool, 1 hook wrench	1.25255.0001
Chromolith® Prep end cap set		25 mm			1 inlet cap complete,1 outlet cap	1.25256.0001
Chromolith® Prep frit set		25 mm			10 frits	1.25257.0001
Chromolith® Prep 25 mm guard cartridge holder		25 mm	10 mm		1 cartridge holder	1.25262.0001
Chromolith® Prep 25 mm column coupler		25 mm			1 column coupler	1.25259.0001

* Guard column type examples and detailed information please find on page 34.

For further information on Merck Millipore
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