MACHEREY-NAGEL

HPLC Columns

Adsorbents for Liquid Chromatography

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Columns for HPLC

Adsorbents for liquid chromatography

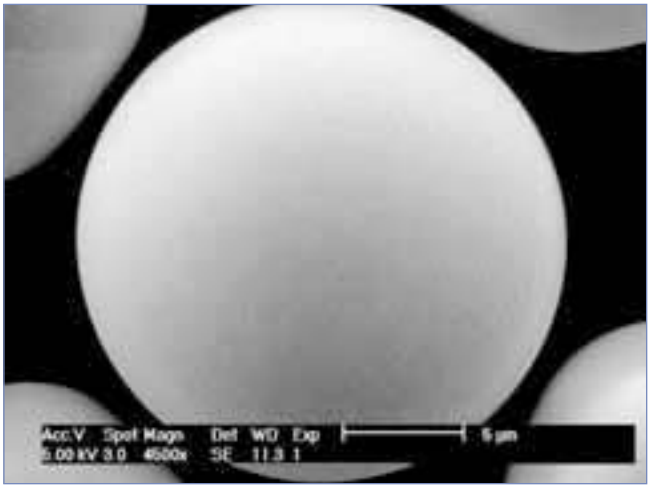
NUCLEODUR® high purity silica for HPLC

NUCLEODUR® is a fully synthetical type B silica (silica of 3rd generation) offering highly advanced physical properties like **totally spherical** particle shape, outstanding **surface microstructure**, high **pressure stability** and **low metal content**.

NUCLEODUR® as a state-of-the-art silica is the ideal base material for modern HPLC phases. It is the result of MACHEREY-NAGEL's pioneering research in chromatography for more than 40 years and succeeds MN's famous NUCLEOSIL® silica.

In RP liquid chromatography the efficiency of the packing is strongly affected by the quality of the base silica itself. Shortcomings in the surface geometry of the particles or metal contaminants are the main reasons for inadequate coverage with the covalently bonded alkylsilanes in the subsequent derivatization steps. It is well known, that poor surface coverage and, in consequence, high activity of residual free silanols often results in peak tailing or adsorption, particularly with basic compounds.

Particle shape and surface symmetry



NUCLEODUR® silicas are synthesized in a unique and carefully controlled manufacturing process which provides silica particles, which are totally spherical. The picture shows the outstanding smoothness of the NUCLEODUR® surface.

Purity

As already mentioned above, a highly pure silica is required for achieving symmetric peak shapes and maximum resolution. Inclusions of e.g. iron or alkaline earth metal ions on the silica surface are largely responsible for the unwanted interactions with ionizable analytes, e.g. amines or phenolic compounds.

NUCLEODUR® is virtually free of metal impurities and low acidic surface silanols. Elemental analysis data of NUCLEODUR® 5 µm measured by AAS are listed below.

Elementary analysis (metal ions) of NUCLEODUR® 100-5

Aluminium	<	5	ppm
Iron	<	5	ppm
Sodium	<	5	ppm
Calcium	<	10	ppm
Titanium	<	1	ppm
Zirconium	<	1	ppm
Arsenic	<	0.5	ppm
Mercury	<	0.05	ppm

Pressure stability

The totally spherical and 100% synthetic silica gel exhibits an outstanding mechanical stability, even at high pressures up to 800 bar and elevated eluent flow rates.

In addition, after several cycles of repeated packing, no significant drop in pressure can be observed. The latter is of prime importance for preparative and process-scale applications.

Physical data of NUCLEODUR®

Surface area (BET)	340 m^2/g
Pore size	110 Å
Pore volume	0.9 ml/g

NUCLEODUR® modifications

Several different surface modifications based on NUCLEODUR® silica have been developed over the last years providing a full range of specified HPLC phases and an ideal tool for every separation:

- NUCLEODUR® C_{18} Gravity and C_8 Gravity
- NUCLEODUR® C_{18} Isis
- NUCLEODUR® C_{18} Pyramid
- NUCLEODUR® Sphinx RP
- NUCLEODUR® C_{18} HTec *NEW!*
- NUCLEODUR® C_{18} ec and C_8 ec
- NUCLEODUR® HILIC *NEW!*
- NUCLEODUR® CN and CN-RP
- NUCLEODUR® NH_2 and NH_2-RP *NEW!*

For a summary of important properties of our NUCLEODUR® phases please see page 102.

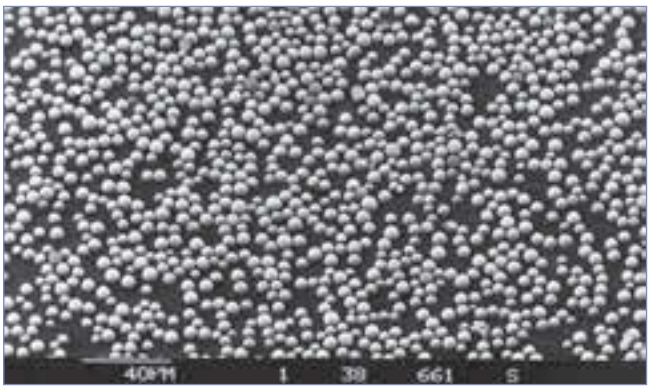
NUCLEOSIL® standard silica for HPLC

NUCLEOSIL® is a family of totally porous spherical silicas. They feature a very pure and uniform SiO_2 structure and have gained wide acceptance as routine chromatographic packings for very different fields of modern chromatography.

- one of the first spherical silicas used in HPLC
- developed in the early seventies, it became a world-renowned HPLC packing
- still found in many analytical and preparative applications, it is an absolutely reliable choice in HPLC
- the largest variety of modified HPLC silicas available

Due to its particle sizes NUCLEOSIL® finds application in analytical as well as in preparative columns. It allows

- high bed stability due to spherical particles
- high efficiency due to narrow particle size distribution
- high separation performance due to optimized binding techniques
- high chemical and mechanical stability
- high load capacity and recovery rates
- high reproducibility from lot to lot



Physical properties of NUCLEOSIL® silicas

NUCLEOSIL® is manufactured with different pore diameters (50, 100, 120, 300, 500, 1000 and 4000 Å) and particle sizes from 3 µm (only NUCLEOSIL® 50, 100 and 120) to 10 µm with very narrow fractionation.

All narrow-pore NUCLEOSIL® packings are stable up to 600 bar (8 500 psi), for NUCLEOSIL® 120 even pressures of up to 800 bar (11 500 psi) can be applied. The wide-pore NUCLEOSIL® silicas are stable up to 300 or 400 bar (4 200 or 5 600 psi).

For a summary of physical properties of unmodified NUCLEOSIL® silica see page 140.

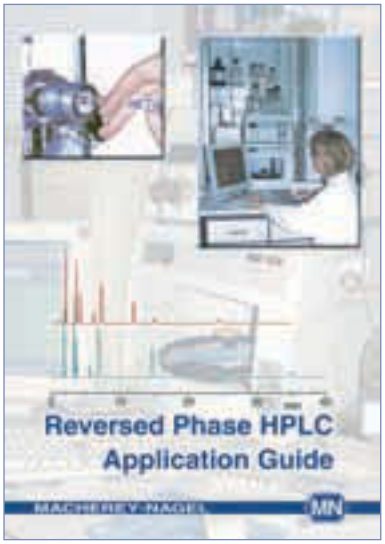
NUCLEOSIL® modifications

NUCLEOSIL® packings are available as unmodified silica or with numerous chemically bonded phases:

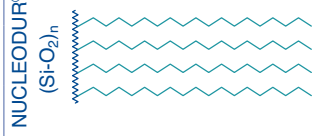
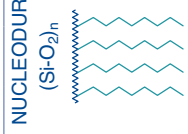
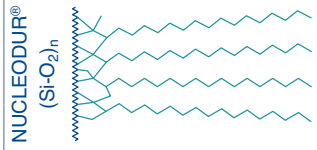
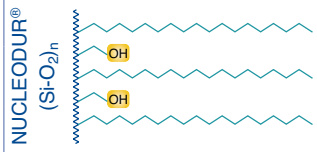
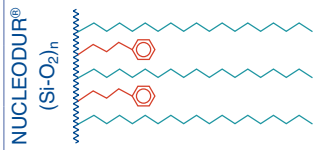
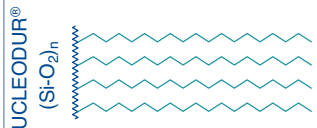
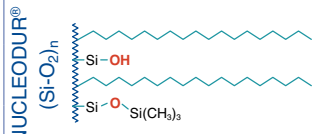
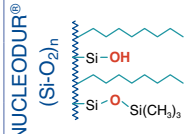
- RP phases like C_{18} AB, C_{18} HD, C_{18} Nautilus, C_{18} endcapped, Protect I, C_8 HD, C_8 ec, C_8, C_4, C_2 and Phenyl) separate mainly by hydrophobic interactions (van der Waals forces).
  The less polar the sample molecules, the more they are retained – the more polar the sample, the weaker are the hydrophobic interactions and consequently the shorter are retention times.
- Phases with chemically bonded polar groups such as CN, NO_2, NH_2, $N(CH_3)_2$, OH show selective separation properties.
  Due to the availability of different functional groups it is possible to vary the chemical characteristics of the surface and consequently the adsorption characteristics of the stationary phase.
- Silica-based ion exchangers (NUCLEOSIL® SA and SB) are stable from pH 2 to 8 and do not swell. Compared to resin-based ion exchangers they offer the advantage of constant permeability, even when the ionic strength and/or pH of the eluent are changed. The separation can be influenced by
  – the **type of buffer**
  – the **ionic strength** and
  – the **pH value.**

For a summary of our NUCLEOSIL® phases please refer to page 130.

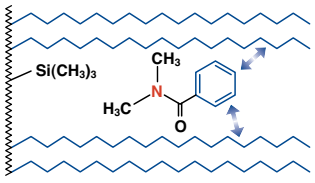
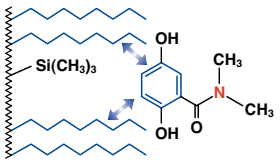
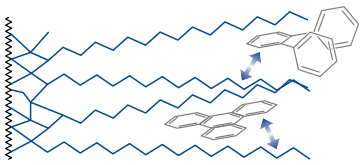
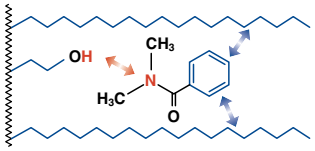
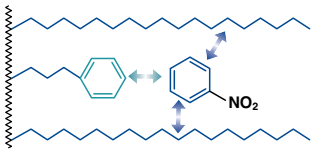
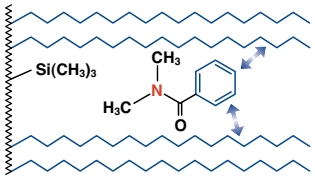
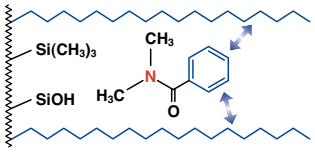
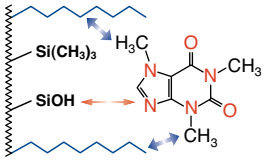
For basic information on RP chromatography and numerous applications with our NUCLEODUR® and NUCLEOSIL® phases please ask for our Reversed Phase HPLC Application Guide.



Overview of NUCLEODUR® RP phases

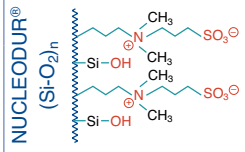
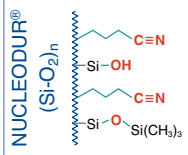
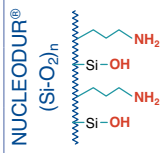
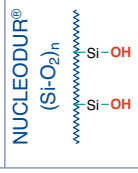
Phase	Specification	Characteristics*	Stability	Structure
C18 Gravity	octadecyl phase, high density coating multi-endcapping 18 % C · USP **L1**	A B C	pH stability 1 – 11, suitable for LC/MS	
C8 Gravity	octyl phase, high density coating multi-endcapping 11 % C · USP **L7**	A B C	pH stability 1 – 11, suitable for LC/MS	
C18 Isis	octadecyl phase with specially crosslinked surface modification endcapping 20 % C · USP **L1**	A B C	pH stability 1 – 10, suitable for LC/MS	
C18 Pyramid	C_{18} modification with polar endcapping 14 % C · USP **L1**	A B C	stable against 100 % aqueous eluents, pH stability 1 – 9, suitable for LC/MS	
Sphinx RP	bifunctional RP phase, propylphenyl and C_{18} ligands; endcapping 15 % C; USP **L1** and **L11**	A B C	pH stability 1 – 10, suitable for LC/MS	
C18 HTec	octadecyl phase with high capacity, high density coating, multi-endcapping 18 % C · USP **L1**	A B B	pH stability 1 – 11, suitable for LC/MS	
C18 ec	octadecyl / octyl phase, medium density coating endcapping 17.5 % C · USP **L1**	A B C	pH stability 1 – 9	
C8 ec	octadecyl / octyl phase, medium density coating endcapping 10.5 % C · USP **L7**	A B C	pH stability 1 – 9	

* A = hydrophobic selectivity, B = polar/ionic selectivity, C = steric selectivity

	Application	Similar phases**	Separation principle · Retention mechanism		Page
	in general compounds with ionizable functional groups such as basic pharmaceuticals and pesticides	**NUCLEOSIL® C_{18} HD** Waters Xterra® RP_{18} / MS C_{18}; Phenomenex Luna® C18 (2), Gemini®, Synergi™ and Max RP; Zorbax Extend C18; Inertsil ODS III; Purospher RP-18; Star RP-18	**only hydrophobic interactions** (van der Waals interactions)		108 – 111
	like C_{18} Gravity, however generally shorter retention times for nonpolar compounds	**NUCLEOSIL® C_8 HD** Waters Xterra® RP_8 / MS C_8; Phenomenex Luna C8; Zorbax Eclipse; XDB-C8			
	high steric selectivity, thus suited for separation of positional and structural isomers, planar/nonplanar molecules	**NUCLEOSIL® C_{18} AB** Inertsil ODS-P; YMC Pro C18RS, Zorbax SB	**steric interactions** and **hydrophobic interactions**		112 – 113
	basic pharmaceutical ingredients, very polar compounds, organic acids	Phenomenex Aqua; YMC AQ; Waters Atlantis® dC18	**hydrophobic interactions** and **polar interactions** (H bonds)		114 – 115
	compounds with aromatic and multiple bond systems	no similar phases	**π-π interactions** and **hydrophobic interactions**		116 – 117
	robust and well base deactivated C_{18} phase; all separation tasks with preparative potential	Waters Xterra® RP_{18} / MS C_{18} / SunFire™ C_{18}; Phenomenex Luna® C18 (2), Gemini®, Synergi™ and Max RP; Zorbax Extend C18; Inertsil ODS III; Purospher RP-18; Star RP-18	**only hydrophobic interactions** (van der Waals interactions)		118 – 119
	robust C_{18} phase for routine analyses	**NUCLEOSIL® C_{18}** Spherisorb® ODS II; Hypersil ODS; Waters Symmetry® C18; Inertsil ODS II; Kromasil C18; LiChrospher RP 18	**only hydrophobic interactions** (van der Waals interactions) some residual silanol interactions		120 – 122
	robust C_8 phase for routine analyses	**NUCLEOSIL® C_8 ec / C_8** Spherisorb® C8; Hypersil MOS; Waters Symmetry® C8; Kromasil C8; LiChrospher RP 8			

** phases which provide a similar selectivity based on chemical and physical properties

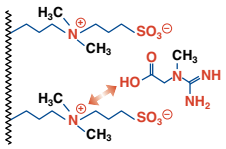
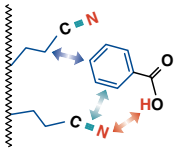
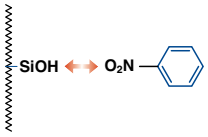
Overview of polar NUCLEODUR® phases

Phase	Specification	Characteristics*		Stability	Structure
HILIC	zwitterionic ammonium sulfonic acid modification 7 % C	A	⬢	pH stability 2 – 8.5, suitable for LC/MS	
		B	⬢⬢⬢⬢⬢		
		C	–		
CN / CN-RP	cyano (nitrile) phase for NP and RP separations 7 % C · USP **L10**	A	⬢	pH stability 1 – 8, stable towards highly aqueous mobile phases	
		B	⬢⬢⬢⬢		
		C	–		
NH_2 / NH_2-RP	amino phase for NP and RP separations 2.5 % C · USP **L8**	A	⬢	pH stability 1 – 8, stable towards highly aqueous mobile phases	
		B	⬢⬢⬢⬢		
		C	–		
SiOH	unmodified high purity silica USP **L3**	A	–	pH stability 2 – 8	
		B	n.a.		
		C	–		

* A = ⬢ hydrophobic selectivity, B = ⬢ polar/ionic selectivity, C = ⬢ steric selectivity

NUCLEODUR® high purity silica

Special selectivities for special separations

	Application	Similar phases**	Separation principle · Retention mechanism		Page
	hydrophilic compounds such as organic polar acids and bases, polar natural compounds	Merck Sequant ZIC®-HILIC; Sielc Obelisc™	ionic / hydrophilic interactions, electrostatic interactions		124 – 125
	polar organic compounds (basic drugs), molecules containing π electron systems	NUCLEOSIL® CN / CN-RP	π-π and polar interactions (H bonds), hydrophobic interactions		126 – 127
	sugars, sugar alcohols and other hydroxy compounds, DNA bases, polar compounds in general	NUCLEOSIL® NH_2 / NH_2-RP	polar /ionic interactions, hydrophobic interactions		128 – 129
	polar compounds in general	unmodified NUCLEOSIL®	polar /ionic interactions		123
		** phases which provide a similar selectivity based on chemical and physical properties			

Columns for HPLC

An optimised phase for every field of application

Particle size and separation efficiency

1.8 µm particles for increased separation efficiency

- **key features**
  - decrease of analysis time (ultra fast HPLC)
  - shorter columns with high separation efficiency
  - significant improvement of resolution and detection sensitivity
  - suitable for **LC/MS** due to low bleeding characteristics
- NUCLEODUR® 1.8 µm particles are fractionated to limit the increase in back pressure

- **NUCLEODUR® phases available in 1.8 µm:**
  C_{18} Gravity
  C_8 Gravity
  C_{18} Isis
  C_{18} Pyramid
  Sphinx RP
  HILIC

Advantages of 1.8 µm particle size

Miniaturization in HPLC has a long history. It started in the early stage of HPLC development with the reduction of particle size from 10 µm via 7 µm to standard 5 µm – which is still the most widely used particle diameter in analytical HPLC – to 3 µm spherical particles which so far was the smallest particle size available for gaining higher theoretical plates and efficiencies. With the introduction of the new 1.8 µm NUCLEODUR® particles now researchers have turned over a new leaf in HPLC column technology. Columns packed with these sub-2 micron particles show extraordinary improvements in terms of plate numbers, column efficiencies and resolution compared with their 3 µm counterparts.

Features of 1.8 µm NUCLEODUR® silica particles

- **Increase of separation efficiency by higher number of theoretical plates (N):**
  50 x 4.6 mm NUCLEODUR® C_{18} Gravity
  3 µm: N ≥ 100 000 plates/m (h value ≤ 10)
  1.8 µm: N ≥ 166 667 plates/m (h value ≤ 6)

  Increase of the plate number by ~67% offers the possibility of using shorter columns with equal plate numbers resulting in a decrease of analysis time.

- **Significant improvement in resolution**

  Use of 1.8 µm instead of 3 µm particles leads to an increase of resolution by a factor 1.29 (29%) since the resolution is inversely proportional to the square root of the particle size:

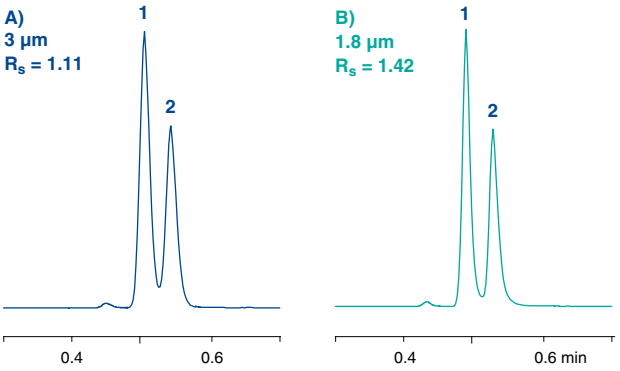
$$R_s = \frac{\sqrt{N}}{4}\left(\frac{\alpha-1}{\alpha}\right)\left(\frac{k_i'}{k_i'+1}\right)$$

R_s = resolution
α = selectivity (separation factor)
k_i = retention
N = plate number with $N \propto 1/d_P$
d_P = particle size

Resolution as a function of particle size

Column: 50 x 4 mm NUCLEODUR® C_{18} Gravity
A) 3 µm, B) 1.8 µm
Eluent: acetonitrile – water (80:20, v/v)
Flow rate: 2 ml/min
Pressure: A) 80 bar, B) 160 bar
Detection: UV, 254 nm

Peaks:
1. Naphthalene
2. Ethylbenzene



- **Column back pressure**

  Due to the smaller particle size the back pressure will increase according to

$$\Delta_P = \frac{\Phi \cdot L_C \cdot \eta \cdot u}{d_P^2}$$

Δ_P = pressure drop
Φ = flow resistance (nondimensional)
L_C = column length
η = viscosity
u = linear velocity
d_P = particle diameter

Because of the high sphericity of the NUCLEODUR® particles and the very narrow particle size distribution we were able to keep the back pressure on a moderate level. Nevertheless the use of columns packed with sub 2 µm particles generally makes special demands on the HPLC equipment. Pumps should be designed for pressures of 250 – 1000 bar and the entire system should feature the lowest possible dead volume.

Particle size and separation efficiency

Comparison of back pressures:		
Eluent:	100 % methanol	
Flow rate:	1.5 ml/min	
Temperature:	22 °C	
Column dimension:	50 x 4.6 mm	
	NUCLEODUR® C_{18} Gravity	Competitor A
3 µm	70 bar	–
1.8 µm	130 bar	170 bar

Technical requirements

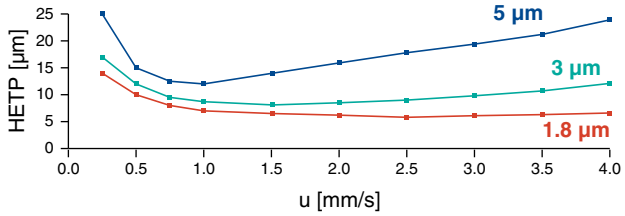
To gain the best result in ultra fast HPLC based on 1.8 µm particles certain technical demands on the instrument are made. Pumps for pressures of 250 – 1000 bar realizing a flow rate of 2 – 3 ml are required. The dead volume of the LC system has to be reduced to a minimum. In addition, fast data recording is necessary for an optimum chromatographic result.

Higher flow rates and shorter run times

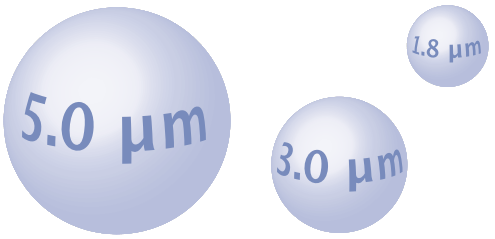
optimal flow rate for 1.8 µm particles is higher than for 3 and 5 µm particles (see figures – the flow rate should be at the van-Deemter minimum)

Van-Deemter plot

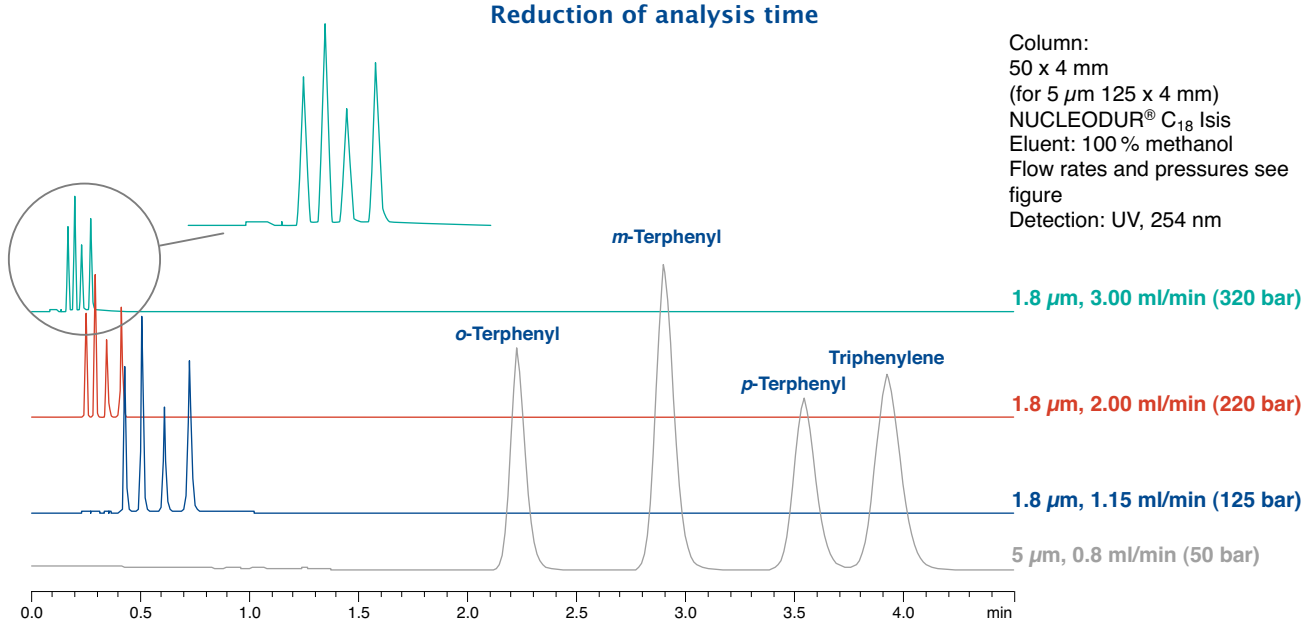
column 50 x 4.6 mm, acetonitrile – water (50:50, v/v), analyte toluene



Currently all NUCLEODUR® premium phases (C_{18} Gravity, C_8 Gravity, C_{18} Isis, C_{18} Pyramid, Sphinx RP, HILIC) are available in 1.8 µm. The description of each phase and its selectivity can be found in the individual chapters.



Reduction of analysis time



NUCLEODUR® C_{18} Gravity · C_8 Gravity — nonpolar high density phases

- **key features:**
  - suitable for **LC/MS** and HPLC at pH extremes (pH 1 – 11)
  - superior base deactivation
  - ideal for method development
- **technical characteristics:**

  available as octadecyl (C_{18}) and octyl (C_8)

  pore size 110 Å; particle sizes 1.8 µm, 3 µm and 5 µm for C_{18}, 1.8 and 5 µm for C_8; 7, 10, 12 and 16 µm particles for preparative purposes on request

  carbon content 18 % for C_{18}, 11 % for C_8
- **recommended application:**

  overall sophisticated analytical separations

  compound classes separated so far: pharmaceuticals, e.g. analgesics, antiinflammatory drugs, antidepressants; herbicides; phytopharmaceuticals; immunosuppressants

  USP L1 (C_{18}) / USP L7 (C_8)

Base deactivation

NUCLEODUR® C_{18} Gravity and NUCLEODUR® C_8 Gravity are based on the ultrapure NUCLEODUR® silica, which is described above.

A unique derivatization process generates a homogeneous surface with a high density of bonded silanes (carbon content ~18% for C_{18}, ~11% for C_8). The following thorough endcapping suppresses any unwanted polar interactions between the silica surface and the sample, which makes “Gravity” particularly suitable for the separation of basic and other ionizable analytes. Even strongly basic pharmaceuticals like amitriptyline are eluted without tailing under isocratic conditions. For a discussion of the different retention behaviour of octadecyl phases compared to octyl phases see page 121.

Enhanced pH stability

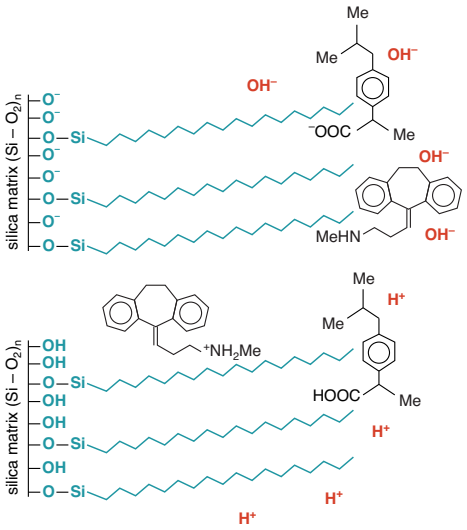
One major disadvantage of using silica stationary phases is the limited stability at strongly acidic or basic pH ranges. Cleavage of the siloxane bonding by hydrolysis, or dissolution of the silica will rapidly lead to a considerable loss in column performance. Therefore conventional RP phases are usually not recommended to be run with mobile phases at pH > 8 or pH < 2 for extended periods of time. The special surface bonding technology and the low concentration of trace elements of NUCLEODUR® C_8 and C_{18} Gravity allow for use at an expanded pH range from pH 1 to 11.

When is enhanced pH stability beneficial?

The option to work at an expanded pH range is often required in method development. Many nitrogen containing compounds like basic drugs are protonated at acidic or neutral pH and exhibit poor retention on a standard C_{18} phase.

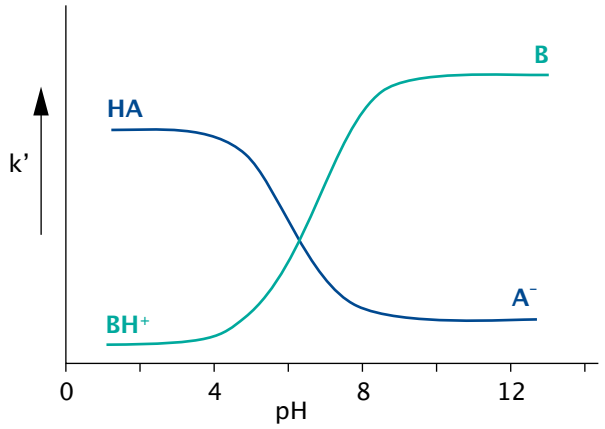
The retention behaviour can be improved by working at a higher pH, where the analyte is no longer protonated, but formally neutrally charged, as a rule between pH 9 – 10. For acidic analytes it is exactly in inverse proportion, maximum retention can be attained at low pH.

Surface silanols at different pH values



The figure above shows the extent of protonation of surface silanols and of two exemplary analytes at acidic and alkaline pH. The following graph explains the general correlation between retention and pH.

Correlation between retention and pH for basic and acidic compounds

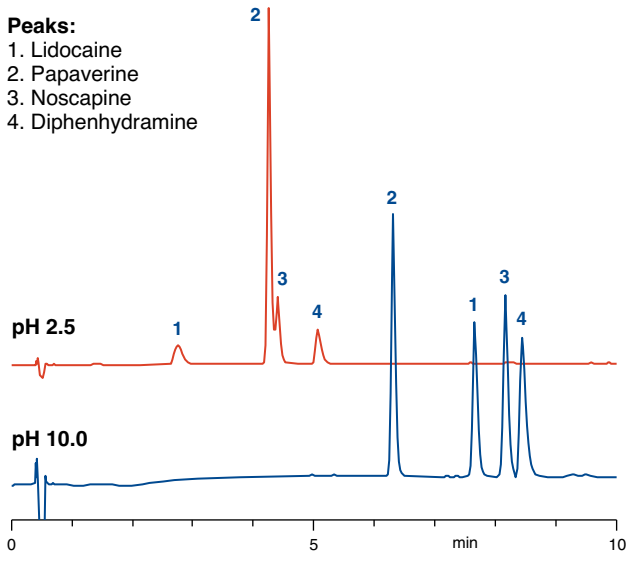


As it was previously mentioned, pH stability of the stationary phase can be helpful for improving selectivity in method development. The figure below shows the separation of 4 basic drugs under acidic and basic conditions.

Separation of basic alkaloids

Column: 125 x 4 mm NUCLEODUR® C_{18} Gravity, 5 µm
Eluents: A) acetonitrile
B) 20 mM $(NH_4)_2HPO_4$, pH 2.5 / 10.0
Gradient: 10% A (1 min) → 75% A in 10 min
Flow rate: 1.0 ml/min
Temperature: 25 °C
Detection: UV, 254 nm
Injection volume: 2 µl

Peaks:
1. Lidocaine
2. Papaverine
3. Noscapine
4. Diphenhydramine



MN Appl. No. 118010

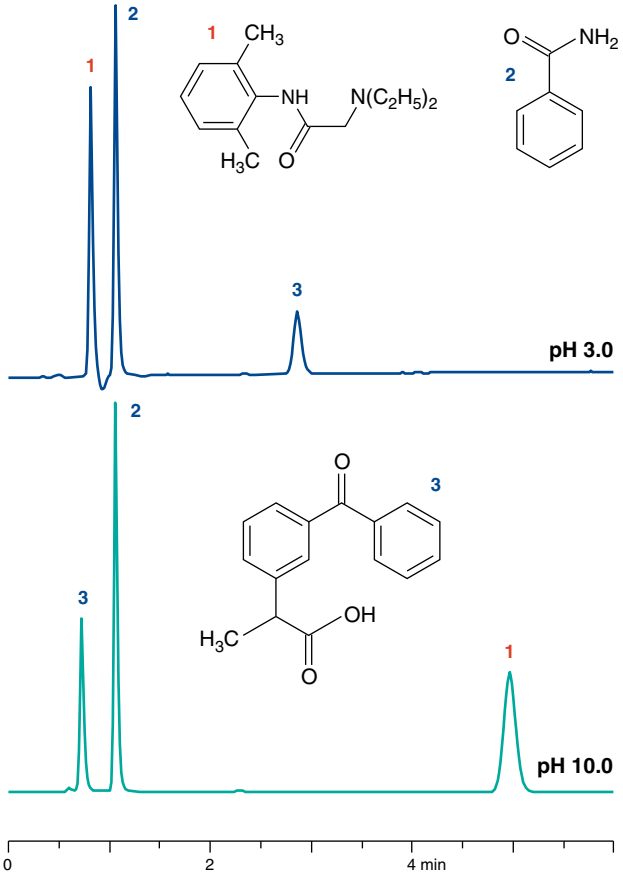
At pH 2.5 the protonated analytes exhibit poor retention (early elution) and in addition an inadequate resolution for papaverine and noscapine, whilst the formally non ionized molecules can be baseline separated due to the better retention pattern at alkaline pH.

A further example how selectivity can be controlled by the pH value is demonstrated below. The sample mixture consists of an acid (ketoprofen), a base (lidocaine) and benzamide. Under acidic conditions the protonated lidocaine is eluted very fast due to lack of sufficiently strong hydrophobic interactions between analyte and C_{18} chains, in contrary to the formally neutral ketoprofen, which is eluted after about 3 minutes. However at pH 10 a reversal of the elution order, with a visibly longer retention time for the basic lidocaine, can be achieved.

Influence of the pH value on selectivity

Column: 125 x 4 mm NUCLEODUR® C_{18} Gravity, 5 µm
Eluents: A) acetonitrile – 10 mM ammonium formate, pH 3.0 (50:50, v/v)
B) acetonitrile – 10 mM ammonium bicarbonate, pH 10.0 (50:50, v/v)
Flow rate: 1.0 ml/min
Temperature: 30 °C
Detection: UV, 230 nm
Injection volume: 2 µl

Peaks:
1. Lidocaine
2. Benzamide
3. Ketoprofen



MN Appl. No. 120860

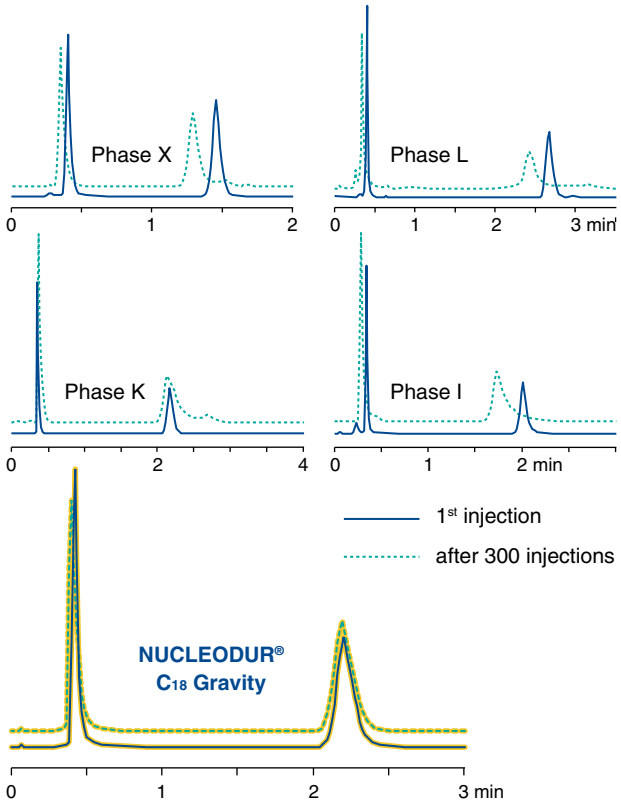
Columns with NUCLEODUR® phases

The following chromatograms demonstrate the stability of NUCLEODUR® C_{18} Gravity under alkaline conditions in comparison with four commercially available modern RP18 phases. Again, the ultrapure Gravity with its unique high density surface bonding technology withstands strong alkaline mobile phase conditions.

Stability of NUCLEODUR® C_{18} Gravity at alkaline pH compared with different C_{18} phases

Columns: 50 x 4.6 mm
Eluent: methanol – water – ammonia (20:80:0.5, v/v/v), pH 11
Flow rate: 1.3 ml/min
Temperature: 30 °C
Detection: UV, 254 nm
Injection volume: 2.0 µl

Peaks: 1. theophylline, 2. caffeine



MN Appl. No. 120850

Even after 300 injections no loss of column efficiency, identified e.g. by peak broadening or decrease in retention times, could be observed.

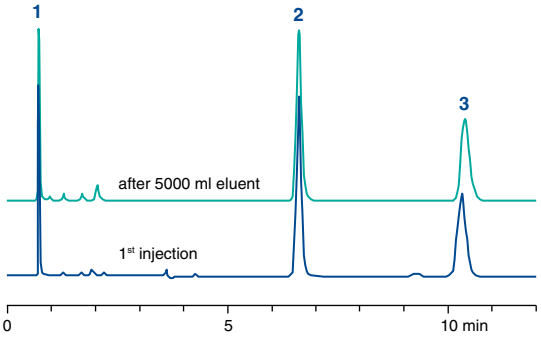
The pH stability of silica under alkaline conditions is mainly a kinetic effect and based on the velocity of the dissolution of the silica support. It is worth mentioning, that this phenomenon also depends on type and concentration of buffers, as well as on the temperature. It is well known that the use of phosphate buffers, particularly at elevated temperatures, can reduce column lifetime even at moderate pH. If possible, phosphate buffers should be replaced by less harmful alternatives.

The following chromatograms show the excellent column stability of NUCLEODUR® C_{18} Gravity in acidic conditions. The retention time of all three compounds in the column performance test remains consistent and virtually unchanged, even after the column is run with 5000 ml eluent. Due to the extremely stable surface modification, no cleavage of the Si–O–Si bonding occurs, column deterioration is therefore successfully prevented.

Stability of NUCLEODUR® C_{18} Gravity at pH 1.5

Column: 125 x 4 mm NUCLEODUR® C_{18} Gravity, 5 µm
Eluent: acetonitrile – 1% TFA in water (50:50, v/v), pH 1.5
Flow rate: 1.0 ml/min
Temperature: 30 °C,
Detection: UV, 230 nm
Injection volume: 5 µl

Peaks: 1. pyridine, 2. toluene, 3. ethylbenzene



MN Appl. No. 120840

Ordering information

eluent in column acetonitrile / water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® C_{18} Gravity, 1,8 µm						octadecyl phase, particle size 1.8 µm, 18% C		
EC columns								
2 mm ID	**760078.20**	**760079.20**	**760071.20**	**760076.20**		**760075.20**		
3 mm ID	**760078.30**	**760079.30**						
4 mm ID	**760078.40**	**760079.40**						
4.6 mm ID	**760078.46**	**760079.46**						

Columns with NUCLEODUR® phases

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® C_{18} Gravity, 3 µm					octadecyl phase, particle size 3 µm, 18 % C			
Microbore columns								
1 mm ID				717714.10	717715.10	717716.10	717717.10	
EC columns								
2 mm ID		760080.20			760081.20	760083.20	760082.20	761124.30
3 mm ID		760080.30			760081.30	760083.30	760082.30	761124.30
4 mm ID		760080.40			760081.40	760083.40	760082.40	761124.40
4.6 mm ID		760080.46	760086.46	760084.46	760081.46	760083.46	760082.46	761124.40
NUCLEODUR® C_{18} Gravity, 5 µm					octadecyl phase, particle size 5 µm, 18 % C			
Microbore columns								
1 mm ID				717706.10	717707.10	717708.10	717705.10	
EC columns								
2 mm ID		760102.20			760100.20	760103.20	760101.20	761125.30
3 mm ID		760102.30			760100.30	760103.30	760101.30	761125.30
4 mm ID		760102.40			760100.40	760103.40	760101.40	761125.40
4.6 mm ID		760102.46	760106.46	760104.46	760100.46	760103.46	760101.46	761125.40
VarioPrep columns								
10 mm ID		762103.100			762109.100		762113.100	762160.80
21 mm ID		762103.210			762109.210		762113.210	762161.160
32 mm ID							762113.320	762163.320
40 mm ID						762100.400	762113.400	762163.320
NUCLEODUR® C_{18} Gravity, 10 µm					octadecyl phase, particle size 10 µm, 18 % C			
VarioPrep columns								
21 mm ID							762250.210	762161.160
40 mm ID							762250.400	762163.320
NUCLEODUR® C_8 Gravity, 1.8 µm					octyl phase, particle size 1.8 µm, 11 % C			
EC columns								
2 mm ID	760756.20	760755.20	760760.20	760757.20		760759.20		
3 mm ID	760756.30	760755.30						
4 mm ID	760756.40	760755.40						
4.6 mm ID	760756.46	760755.46						
NUCLEODUR® C_8 Gravity, 5 µm					octyl phase, particle size 5 µm, 11 % C			
EC columns								
2 mm ID		760750.20			760751.20	760752.20	760753.20	761754.30
3 mm ID		760750.30			760751.30	760752.30	760753.30	761754.30
4 mm ID		760750.40			760751.40	760752.40	760753.40	761754.40
4.6 mm ID		760750.46	760749.46	760754.46	760751.46	760752.46	760753.46	761754.40
VarioPrep columns								
10 mm ID		762081.100			762071.100		762070.100	762097.80
21 mm ID		762081.210			762071.210	762082.210	762070.210	762089.160

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, analytical and preparative main columns in packs of 1.
Other Microbore and VarioPrep columns on request; for available dimensions see page 168/169.

VP guard column cartridge	pack of	for VP column	required holder (see page 168)
VP 10/8, 10 x 8 mm ID	2	10 mm ID	8 mm (REF 718251)
VP 20/16, 20 x 16 mm ID	2	21 mm ID	16 mm (REF 718250)
VP 15/32, 15 x 32 mm ID	1	32 to 40 mm ID	32 mm (REF 718253)

NUCLEODUR® C_{18} Isis — phase with high steric selectivity

- **key features:**
  - exceptional steric selectivity
  - outstanding surface deactivation
  - suitable for **LC/MS** and HPLC at pH 1 – 10
- **technical characteristics:**
  C_{18} phase with special polymeric, crosslinked surface modification; pore size 110 Å; particle sizes 1.8 µm, 3 µm and 5 µm; carbon content 20 %
- **recommended application:**
  steroids, (*o,p,m*–) substituted aromatics, fat-soluble vitamins
  USP L1

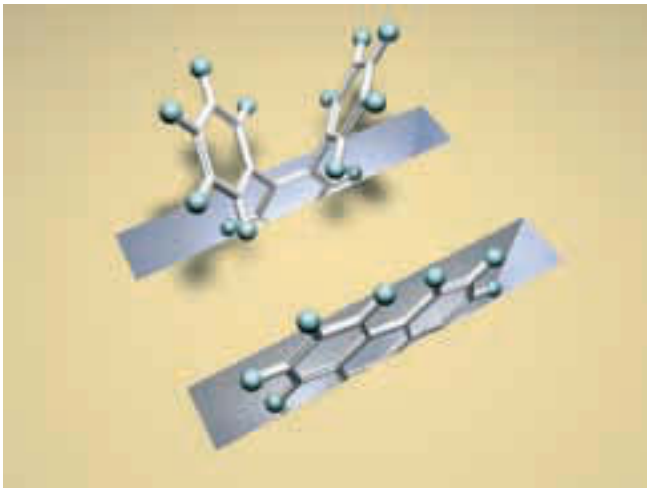
Surface modification

By use of specific C_{18} silanes and appropriate polymeric bonding technologies a dense shield of alkyl chains protects the subjacent silica matrix. Elemental analysis of NUCLEODUR® C_{18} Isis shows a carbon load of 20%.

The target crosslinking of the C_{18} chains on the surface enables the separation of compounds with similar molecular structure but different stereochemical properties. The technical term for this feature is steric selectivity. The chromatograms on the right reveal the improved resolution for positional isomers in a test mixture of aromatic compounds on NUCLEODUR® C_{18} Isis (1) in direct comparison with monomerically coated (2) and polar endcapped (3) C_{18} columns.

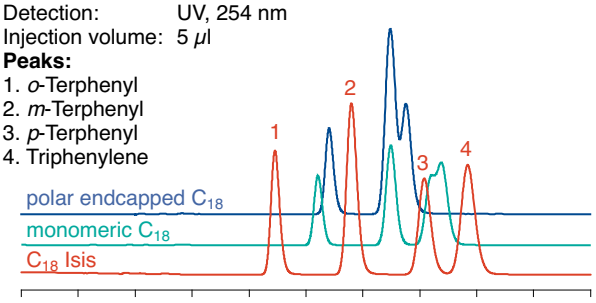
Sander and Wise [LCGC 8 (1990) 378 – 390] proposed a model for the retention of aromatic compounds based on molecular shape, which is referred to as "Slot Model". This model pictures the bonded C_{18} phase on the silica surface with slots which the analytes have to penetrate during retention. Planar molecules are able to penetrate these slots deeper than non-planar molecules of similar molecular weight and length-to-breadth ratio. Thus triphenylene is longer retained than *o*-terphenyl.

Slot model



Steric selectivity of NUCLEODUR® C_{18} Isis

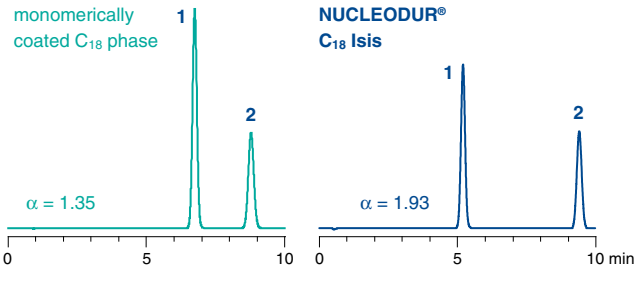
Columns: 125 x 4 mm; **NUCLEODUR® C_{18} Isis, monomerically coated C_{18} phase, polar endcapped phase**
Eluent: methanol – water (90:10, v/v)
Flow rate: 1 ml/min, temperature: 35 °C
Detection: UV, 254 nm
Injection volume: 5 µl
Peaks:
1. *o*-Terphenyl
2. *m*-Terphenyl
3. *p*-Terphenyl
4. Triphenylene



The separation of *o*-terphenyl and triphenylene is a concrete example to evaluate the selectivity potential of a reversed phase column in terms of the different shape of two molecules. The phenyl rings of *o*-terphenyl are twisted out of plane while triphenylene has a planar geometry. The separation factor (α value) is a measure for the steric selectivity. As is shown in the following chromatograms the α value is considerable larger on NUCLEODUR® C_{18} Isis compared to a conventional C_{18} column.

Steric selectivity of NUCLEODUR® C_{18} Isis

Columns: 125 x 4 mm
Eluent: methanol – water (80:20, v/v)
Flow rate: 1 ml/min, temperature: 40 °C
Detection: UV, 254 nm, injection volume: 1 µl
Peaks: 1. *o*-terphenyl, 2. triphenylene



Columns with NUCLEODUR® phases

Surface deactivation

The chromatography of basic analytes requires a high density of surface-bonded C_{18} silanes combined with a thorough endcapping procedure to keep silanol activity at a minimum. This ensures tailing-free elution of even strongly basic amino-containing compounds (see Appl. 121210 at *www.mn-net.com*).

Stability

The applied special surface bonding technology also provides improved stability features for the NUCLEODUR® C_{18} Isis phase.

Ordering information

eluent in column acetonitrile / water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® C_{18} Isis, 1.8 µm								particle size 1.8 µm
EC columns								
2 mm ID	**760406.20**	**760405.20**	**760396.20**	**760407.20**		**760409.20**		
3 mm ID	**760406.30**	**760405.30**						
4 mm ID	**760406.40**	**760405.40**						
4.6 mm ID	**760406.46**	**760405.46**						
NUCLEODUR® C_{18} Isis, 3 µm								particle size 3 µm
Microbore columns								
1 mm ID		**717760.10**		**717761.10**	**717762.10**			
EC columns								
2 mm ID		**760400.20**			**760402.20**	**760403.20**	**760404.20**	**761300.30**
3 mm ID		**760400.30**			**760402.30**	**760403.30**	**760404.30**	**761300.30**
4 mm ID		**760400.40**			**760402.40**	**760403.40**	**760404.40**	**761300.40**
4.6 mm ID		**760400.46**	**760397.46**	**760401.46**	**760402.46**	**760403.46**	**760404.46**	**761300.40**
NUCLEODUR® C_{18} Isis, 5 µm								particle size 5 µm
Microbore columns								
1 mm ID		**717770.10**		**717771.10**	**717772.10**			
EC columns								
2 mm ID		**760410.20**			**760412.20**	**760413.20**	**760414.20**	**761310.30**
3 mm ID		**760410.30**			**760412.30**	**760413.30**	**760414.30**	**761310.30**
4 mm ID		**760410.40**			**760412.40**	**760413.40**	**760414.40**	**761310.40**
4.6 mm ID		**760410.46**	**760416.46**	**760415.46**	**760412.46**	**760413.46**	**760414.46**	**761310.40**
VarioPrep columns								
10 mm ID		**762404.100**			**762405.100**		**762403.100**	**762420.80**
21 mm ID		**762404.210**			**762405.210**		**762403.210**	**762421.160**
32 mm ID							**762403.320**	**762422.320**
40 mm ID						**762406.400**	**762403.400**	**762422.320**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, analytical and preparative main columns in packs of 1.
Other Microbore and VarioPrep columns on request; for available dimensions see page 168/169.

VP guard column cartridge	pack of	for VP column	required holder (see page 168)
VP 10/8, 10 x 8 mm ID	2	10 mm ID	8 mm (REF 718251)
VP 20/16, 20 x 16 mm ID	2	21 mm ID	16 mm (REF 718250)
VP 15/32, 15 x 32 mm ID	1	32 to 40 mm ID	32 mm (REF 718253)

NUCLEODUR® C_{18} Pyramid — phase for highly aqueous eluents

- **key features:**
  - stable in 100% aqueous mobile phase systems
  - interesting polar selectivity features
  - excellent base deactivation; suitable for **LC/MS** due to low bleeding characteristics
- **technical characteristics:**

  special phase with polar endcapping; pore size 110 Å; particle sizes 1.8 µm, 3 µm and 5 µm (7 and 10 µm particles for preparative purposes on request); carbon content 14%; pH stability 1 – 9
- **recommended application:** analgesics, penicillin antibiotics, nucleic acid bases, water-soluble vitamins, complexing agents, organic acids

  USP L1

RP-HPLC with highly aqueous mobile phases

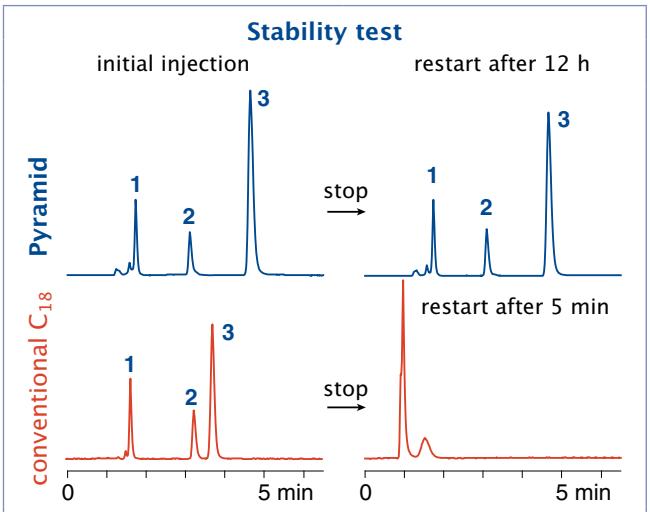
The efforts to neutralize unwanted activity of unreacted surface silanol groups often results in well base-deactivated phases with high carbon load, but a limited scope of selectivity beyond non-polar interactions. In particular polar compounds like carboxylic acids, drug metabolites, etc. show only weak retention on densely bonded reversed phase columns due to distinct hydrophobic properties but low polar interactions. Very polar analytes require highly aqueous mobile phases for solubility and retention. Conventional reversed phase columns often display stability problems in eluent systems with high percentage of water (> 95%) as evidenced by a sudden decrease of retention time and overall poor reproducibility. This phenomenon is described as phase collapse caused by the mobile phase expelled from the pores due to the fact, that hydrophobic RP phases are incompletely wetted with the mobile phase [U. D. Neue et al., Chromatographia 54 (2001) 169 – 177].

Different approaches can be used to increase column stability with highly aqueous mobile phase systems. The most promising concepts are incorporating a polar group in the hydrophobic alkyl chain, or using hydrophilic endcapping procedures to improve the wettability of the reversed phase modification. NUCLEOSIL® Nautilus may be taken as an example for the embedded polar group strategy, in which a C_{18} silane with a polar function is successfully linked to the silica surface [D. Rieger, H. Riering, Int. Laboratory Aug. 2000, Vol. 30 (4A), 12].

Stability features

NUCLEODUR® C_{18} Pyramid is a silica phase with hydrophilic endcapping, designed especially for use in eluent systems of up to 100% water. The upper figure shows the retention behaviour of tartaric, acetic and maleic acid under purely aqueous conditions on NUCLEODUR® C_{18} Pyramid in comparison with a conventionally bonded RP phase.

It can be shown that the retention times for NUCLEODUR® C_{18} Pyramid remain nearly unchanged between initial injection and restart after the flow has been stopped for 12 hours, whilst the performance of the conventional RP column already collapsed totally 5 min.



Both columns: 125 x 4 mm
Eluent: 50 mM KH_2PO_4 pH 2.5
Flow rate: 0.7 ml/min, temperature 25 °C
Detection: UV, 210 nm
Injection volume: 1 µl.
Peaks: 1. tartaric acid, 2. acetic acid, 3. maleic acid

MN Appl. No. 120870

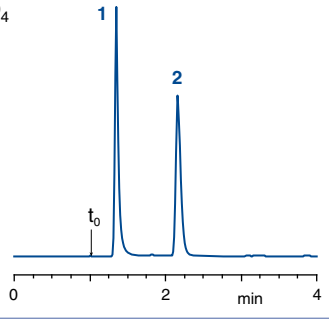
Retention characteristics

The polar surface derivatization exhibits retention characteristics, which differentiate the "Pyramid" from conventional C_{18} stationary phases. The chromatogram below shows the improved retention behaviour of very polar compounds such as short chain organic acids, which are insufficiently retained on RP columns with predominantly hydrophobic surface properties.

Separation of very polar compounds

Column: 125 x 4 mm NUCLEODUR® C_{18} Pyramid, 5 µm
Eluent: 0.2 % H_3PO_4
Flow rate: 1.0 ml/min
Temperature: 22 °C
Detection: UV, 202 nm
Injection volume: 2 µl

Peaks:
1. Formic acid
2. Acetic acid



MN Appl. No. 119170

In addition to the exceptional polar selectivity NUCLEODUR® C_{18} Pyramid also provides adequate hydrophobic retention (see application No. 119180 at *www.mn-net.com*). The capacity factors of the non-polar, alkyl-substituted benzenes toluene and ethylbenzene do not go too far in comparison with standard C_{18} phases.

Base deactivation

The perceptible increase in polarity has no impact on the retention behaviour of ionizable analytes. Even with the strongly basic compounds of the tricyclic antidepressant drug test mixture, no unwanted interactions or a so-called lack in base deactivation are observed (see application 119200 at *www.mn-net.com*).

Ordering information

eluent in column acetonitrile / water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® C_{18} Pyramid, 1.8 µm							particle size 1.8 µm	
EC columns								
2 mm ID	760271.20	760272.20	760275.20	760273.20		760274.20		
3 mm ID	760271.30	760272.30						
4 mm ID	760271.40	760272.40						
4.6 mm ID	760271.46	760272.46						
NUCLEODUR® C_{18} Pyramid, 3 µm							particle size 3 µm	
Microbore columns								
1 mm ID		717740.10		717741.10	717742.10	717743.10	717744.10	
EC columns								
2 mm ID		760263.20			760260.20	760261.20	760262.20	761854.30
3 mm ID		760263.30			760260.30	760261.30	760262.30	761854.30
4 mm ID		760263.40			760260.40	760261.40	760262.40	761854.40
4.6 mm ID		760263.46	760259.46	760264.46	760260.46	760261.46	760262.46	761854.40
NUCLEODUR® C_{18} Pyramid, 5 µm							particle size 5 µm	
Microbore columns								
1 mm ID				717722.10	717723.10	717724.10	717725.10	
EC columns								
2 mm ID		760200.20			760201.20	760203.20	760202.20	761800.30
3 mm ID		760200.30			760201.30	760203.30	760202.30	761800.30
4 mm ID		760200.40			760201.40	760203.40	760202.40	761800.40
4.6 mm ID		760200.46	760205.46	760204.46	760201.46	760203.46	760202.46	761800.40
VarioPrep columns								
10 mm ID		762271.100			762273.100		762272.100	762291.80
21 mm ID		762271.210			762273.210		762272.210	762292.160
32 mm ID							762272.320	762293.320
40 mm ID						762269.400	762272.400	762293.320

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, analytical and preparative main columns in packs of 1.
Other Microbore and VarioPrep columns on request; for available dimensions see page 168/169.

VP guard column cartridge	pack of	for VP column	required holder (see page 168)
VP 10/8, 10 x 8 mm ID	2	10 mm ID	8 mm (REF 718251)
VP 20/16, 20 x 16 mm ID	2	21 mm ID	16 mm (REF 718250)
VP 15/32, 15 x 32 mm ID	1	32 to 40 mm ID	32 mm (REF 718253)

NUCLEODUR® Sphinx RP — bifunctional RP phase

key features:

- distinct selectivity based on well-balanced bifunctional surface coverage
- widens the scope for method development based on additional π–π interactions
- suitable for **LC/MS** due to low bleeding characteristics

technical characteristics:

octadecyl and propylphenyl modified silica; pore size 110 Å; particle sizes 1.8 µm, 3 µm and 5 µm; carbon content 15 %;
pH stability 1 – 10; high reproducibility and consistent quality

recommended application:

quinolone antibiotics, sulfonamides, xanthines, substituted aromatics

USP L1 and L11

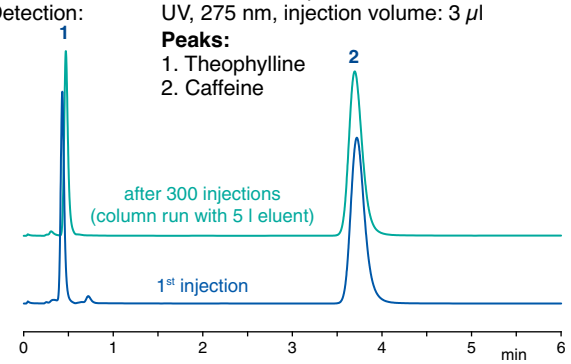
Alternative RP selectivity

NUCLEODUR® Sphinx RP is characterized by exceptional selectivity features generated by a well-balanced ratio of covalently bonded octadecyl and phenyl groups. The combination of classical hydrophobic with π–π interactions (aromatic ring system) expands the scope of selectivity in comparison with conventional reversed phase packings. NUCLEODUR® Sphinx RP is particularly suited for the separation of molecules containing aromatic and multiple bonds. For the separation of polar compounds NUCLEODUR® Sphinx RP can be especially recommended and can also outperform many customary C_{18} phases. In addition, exhaustive endcapping steps minimize unwanted surface silanol activity and guarantee excellent peak shapes even for strong basic analytes.

Stability of NUCLEODUR® Sphinx RP at pH 10

Column: 50 x 4.6 mm NUCLEODUR® Sphinx RP, 5 µm
Eluent: methanol – dil. NH_3, pH 10 (20:80, v/v)
Flow rate: 1.0 ml/min, temperature 30 °C
Detection: UV, 275 nm, injection volume: 3 µl

Peaks:
1. Theophylline
2. Caffeine



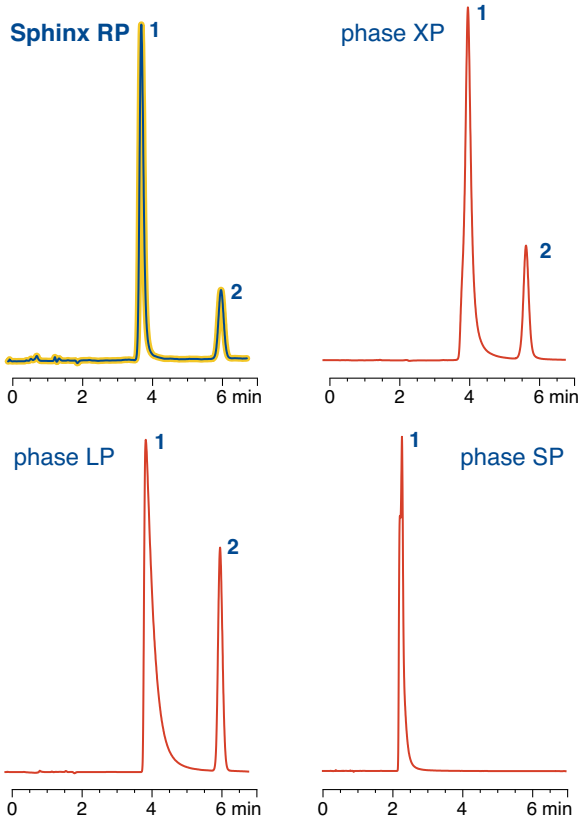
MN Appl. No. 120900

Different from standard phenyl phases, NUCLEODUR® Sphinx RP is far more stable towards hydrolysis and is also suggested for LC/MS applications. Due to the additional intermolecular interactions NUCLEODUR® Sphinx RP is an interesting replenishment to the high density bonded phases NUCLEODUR® C_8/C_{18} Gravity and the polar endcapped NUCLEODUR® C_{18} Pyramid.

Comparison of surface deactivation of different phenyl modified RP phases

Columns: 150 x 4.6 mm, A) NUCLEODUR® Sphinx RP, 5 µm
B) competitor 1 (column XP), C) competitor 2 (column LP)
D) competitor 3 (column SP)
Eluent: methanol – water (30:70, v/v)
Flow rate: 1 ml/min
Temperature: 40 °C
Detection: UV, 254 nm
Injection volume: 2 µl

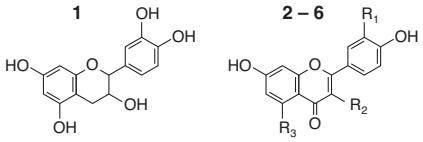
Peaks:
1. Pyridine
2. Phenol



MN Appl. No. 120910

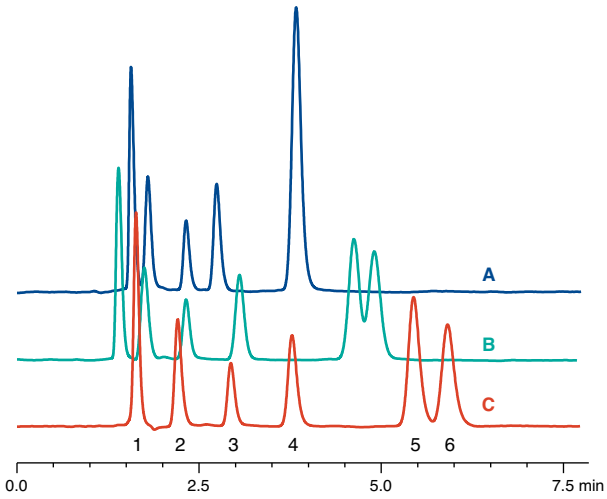
Separation of flavonoids on 3 different NUCLEODUR® phases

Columns: 150 x 4.6 mm: **A) NUCLEODUR® C_8 Gravity, 5 µm**
B) NUCLEODUR® C_{18} Gravity, 5 µm
C) NUCLEODUR® Sphinx RP, 5 µm
Eluent: water – methanol (40:60, v/v), flow rate 1 ml/min
Temperature: 30 °C, detection: UV, 270 nm
Injection volume: 3 µl
Peaks:



1. Catechin
2. Rutin — $R_1 = R_3 = OH$, R_2 = O-rutinose
3. Fisetin — $R_1 = R_2 = OH$, $R_3 = H$
4. Quercetin — $R_1 = R_2 = R_3 = OH$
5. Kaempferol — $R_1 = H$, $R_2 = R_3 = OH$
6. Isorhamnetin — $R_1 = OCH_3$, $R_2 = R_3 = OH$

MN Appl. No. 119830



Ordering information

eluent in column acetonitrile / water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® Sphinx RP, 1.8 µm								particle size 1.8 µm
EC columns								
2 mm ID	760821.20	760822.20	760825.20	760823.20		760824.20		
3 mm ID	760821.30	760822.30						
4 mm ID	760821.40	760822.40						
4.6 mm ID	760821.46	760822.46						
NUCLEODUR® Sphinx RP, 3 µm								particle size 3 µm
Microbore columns								
1 mm ID		717686.10		717685.10		717687.10		
EC columns								
2 mm ID		760806.20			760807.20	760805.20	760808.20	761557.30
3 mm ID		760806.30			760807.30	760805.30	760808.30	761557.30
4 mm ID		760806.40			760807.40	760805.40	760808.40	761557.40
4.6 mm ID		760806.46	760813.46	760812.46	760807.46	760805.46	760808.46	761557.40
NUCLEODUR® Sphinx RP, 5 µm								particle size 5 µm
Microbore columns								
1 mm ID		717680.10		717681.10	717682.10	717683.10	717684.10	
EC columns								
2 mm ID		760800.20			760801.20	760802.20	760803.20	761550.30
3 mm ID		760800.30			760801.30	760802.30	760803.30	761550.30
4 mm ID		760800.40			760801.40	760802.40	760803.40	761550.40
4.6 mm ID		760800.46	760815.46	760809.46	760801.46	760802.46	760803.46	761550.40
VarioPrep columns								
10 mm ID		762372.100			762375.100		762373.100	762390.80
21 mm ID		762372.210			762375.210		762373.210	762391.160
32 mm ID							762373.320	762392.320
40 mm ID						762371.400	762373.400	762392.320

NUCLEODUR® C_{18} HTec — base-deactivated preparative octadecyl phase

key features:

- reliable and durable standard RP phase for up-scaling to preparative scale, suited for **LC/MS**
- high loadability and excellent stability
- outstanding base deactivation

technical characteristics:

high density octadecyl modification (C_{18})
pore size 110 Å; particle sizes 5 µm, 7 µm and 10 µm for analytical and preparative separations
carbon content 18 %, pH stability 1 – 11

recommended application:

sophisticated analytical and preparative separations of basic, neutral and acidic pharmaceuticals, derivatised amino acids, pesticides, fat-soluble vitamins, aldehydes, ketones and phenolic compounds

USP L1

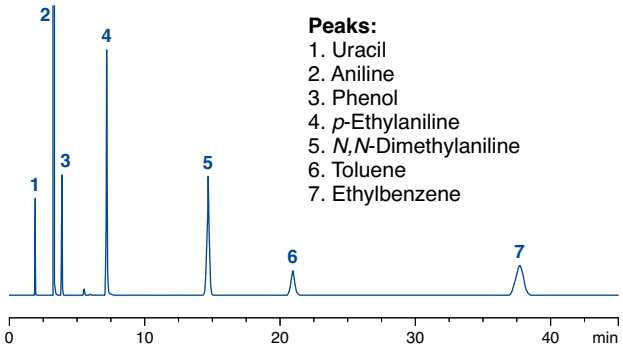
Preparative separations place high demands on silica based HPLC materials. Apart from excellent selectivity and base deactivation, robustness (pH, pressure stability, ...) and capacity are vital criteria for optimal and efficient separation at the preparative scale.

Selectivity and base deactivation

The innovative and special endcapping procedure leads to exceptionally good base deactivation – the Engelhardt test demonstrates superb selectivity, peak symmetry and peak shape over the entire polarity range. In addition NUCLEODUR® C_{18} HTec scores in low bleed characteristics and is therefore highly suitable for LC/MS.

Engelhardt test

Column:	250 x 4 mm NUCLEODUR® 100-5 C_{18} HTec
Eluent:	methanol – water (49:51, v/v)
Flow rate:	1 ml/min
Temperature:	40 °C
Detection:	UV, 254 nm
Injection volume:	5 µl



Peaks:
1. Uracil
2. Aniline
3. Phenol
4. *p*-Ethylaniline
5. *N,N*-Dimethylaniline
6. Toluene
7. Ethylbenzene

MN Appl. No. 123580

Stability and lifetime

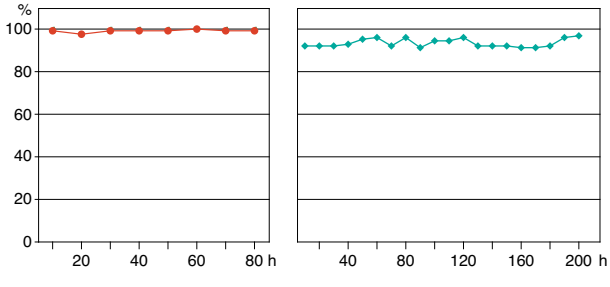
Based on fully synthetic and extremely robust totally spherical NUCLEODUR® silica, NUCLEODUR® C_{18} HTec offers outstanding mechanical rigidity and is thus the perfect choice also for self-packing of prep-columns. The special surface modification and endcapping procedure result in high chemical stability even at extreme chromatographic conditions like high flow rates, temperature or critical solvents (DMSO). Furthermore, NUCLEODUR® C_{18} HTec columns show a remarkably long lifetime in acidic (pH 1) as well as basic (pH 10) mobile phases.

pH stability test

Column:	150 x 4 mm NUCLEODUR® 100-5 C_{18} HTec
Flow rate:	1 ml/min
Detection:	UV, 254 nm
Injection:	5 µl

pH 1:
Eluent: acetonitrile – 1 % TFA in water (50:50, v/v); 80 °C
● % initial retention of ethylbenzene
693 injections

pH 10:
Eluent: methanol – 50 mM triethylamine (25:85, v/v); 50 °C
◆ % initial N of theophylline
1034 injections



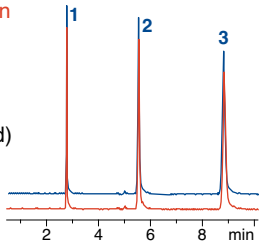
Up-scaling

Due to highest quality standards in our silica production and phase chemistry combined with optimised packing technology, NUCLEODUR® C_{18} HTec delivers exceptional transferability from analytical to preparative scale. This doesn't just apply to the use of different particle sizes (e.g. 5, 7 or 10 µm) but also for diverse column dimensions (e.g. ID 4.6 to 21 mm).

Up-scaling with NUCLEODUR® C_{18} HTec

Columns: **EC 250 x 4.6 mm NUCLEODUR® 100-5 C_{18} HTec**
VP 250 x 21 mm NUCLEODUR® 100-5 C_{18} HTec
Eluent: acetonitrile – water (80:20, v/v)
Flow rates: 1.3 ml/min / 27 ml/min
Temperature: 22 °C
Pressure: 84 bar / 109 bar
Detection: UV, 254 nm
Inj. volume: 3 µl / 60 µl
Peaks: (1 mg/ml of each compound)
1. Phenol
2. Naphthalene
3. Anthracene

MN Appl. No. 123780

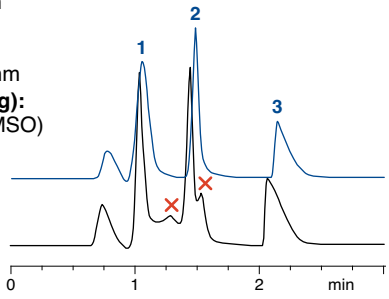


Capacity

A vital criterion for efficiency in preparative HPLC is the capacity of the separation medium. NUCLEODUR® C_{18} HTec is characterised by a notably high loadability under both basic and acidic conditions, while competitor columns show overload effects even at lower loads (x).

Loadability under acidic conditions

Columns: **VP 100 x 21 mm NUCLEODUR® 100-5 C_{18} HTec**
100 x 21.2 mm AXIA™ Gemini® 5 µm C18 110 Å
Eluent: acetonitrile – formic acid in H_2O pH 3.0 (30:70, v/v)
Flow rate: 28 ml/min
Temperature: 22 °C
Pressure: 124 bar
Detection: UV, 254 nm
Peaks (total load 40 mg):
(sample dissolved in DMSO)
1. 4-Acetamidophenol (5 mg)
2. 2-Acetamidophenol (10 mg)
3. Acetylsalicylic acid (25 mg)



Due to innovative surface coating procedures NUCLEODUR® C_{18} HTec offers excellent analytical separation properties and is the first choice for up-scaling to preparative column dimensions.

Ordering information

eluent in column acetonitrile / water

Length →	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® 100-5 C_{18} HTec							particle size 5 µm
EC columns							
2 mm ID	760311.20			760314.20		760316.20	761110.30
3 mm ID	760311.30			760314.30		760316.30	761110.30
4 mm ID	760311.40			760314.40		760316.40	761110.40
4.6 mm ID	760311.46	760312.46	760313.46	760314.46	760315.46	760316.46	761110.40
VarioPrep columns							
10 mm ID	762551.100			762554.100		762556.100	762591.80
21 mm ID	762551.210			762554.210		762556.210	762593.160
32 mm ID						762556.320	762592.320
40 mm ID					762555.400	762556.400	762592.320
50 mm ID						762556.500	762592.320
NUCLEODUR® 100-10 C_{18} HTec							particle size 10 µm
VarioPrep columns							
10 mm ID	762571.100			762574.100		762576.100	762591.80
21 mm ID	762571.210			762574.210		762576.210	762593.160
32 mm ID						762576.320	762592.320
40 mm ID					762575.400	762576.400	762592.320
50 mm ID						762576.500	762592.320

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, analytical and preparative main columns in packs of 1.

NUCLEODUR® HTec bulk material in 5, 7 and 10 µm for self-packing of prep columns see page 172.

VP guard column cartridge	pack of	for VP column	required holder (see page 168)
VP 10/8, 10 x 8 mm ID	2	10 mm ID	8 mm (REF 718251)
VP 20/16, 20 x 16 mm ID	2	21 mm ID	16 mm (REF 718250)
VP 15/32, 15 x 32 mm ID	1	32 to 50 mm ID	32 mm (REF 718253)

NUCLEODUR® C_{18} ec · C_8 ec — nonpolar phases for routine analysis

key features:

- ideal and reliable standard RP phase for daily routine analysis and up-scaling for preparative HPLC
- medium density octadecyl (C_{18}) and octyl (C_8) modification with exhaustive endcapping
- wide range of application areas

technical characteristics:

pore size 110 Å; particle sizes 3 µm and 5 µm; 7 µm, 10 µm, 12 µm, 16 µm, 20 µm, 30 µm and 50 µm for preparative separations; carbon content 17.5 % for C18, 10.5 % for C8

pH stability 1 – 9, high reproducibility from lot to lot

recommended application:

basic, neutral or acidic drugs, derivatised amino acids, pesticides fat-soluble vitamins, aldehydes and ketones, phenolic compounds

USP L1 (C_{18}) / L7 (C_8)

NUCLEODUR® C_{18} ec for daily routine analysis and up-scaling for preparative HPLC

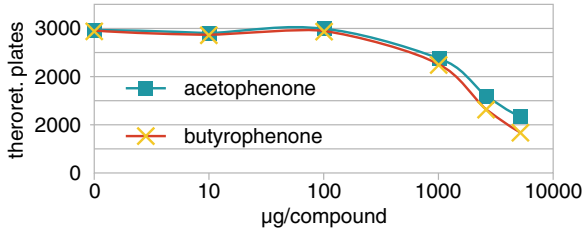
The efficiency of a separation is controlled by particle size and selectivity of the stationary phase. The exceptional surface coverage of monomeric bonded alkylsilanes, combined with an exhaustive endcapping, results in a surface with lowest silanol activity. This allows the tailing-free elution of polar compounds such as basic drugs. NUCLEODUR® C_{18} ec is available in 9 different particle sizes (3, 5, 7, 10, 12, 16, 20, 30 and 50 µm) which cover the whole range from high speed analytical HPLC up to medium and low pressure prep LC. NUCLEODUR® C_{18} ec is also an ideal tool for scale-up purposes.

Loadability

Loadability, probably the most important feature for preparative LC applications, is determined by pore size, pore volume and surface area of the packing. However, it can also be influenced by the molecular weight of the analytes. In the figure below the mass loading curve for acetophenone and butyrophenone on a NUCLEODUR® 100-20 C_{18} ec column describes the correlation between the increase of column loading and the decrease of separation efficiency.

Loading curve

Column: 250 x 4.6 mm NUCLEODUR® 100-20 C_{18} ec, mobile phase: acetonitrile – H_2O 80:20 (v/v), flow: 1.0 ml/min, temperature: 25 °C, detection: UV, 280 – 370 nm



Chemical stability

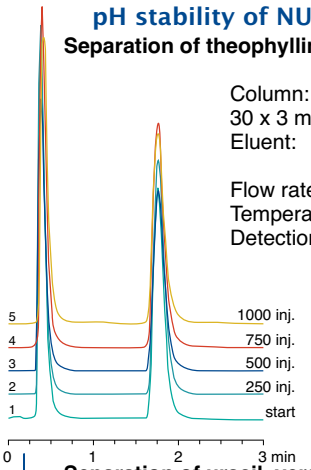
The utmost purity of the base silica and the exceptional silane bonding chemistry minimizes the risk of dissolution, or hydrolysis at pH extremes.

The chromatograms show the retention behavior at pH values of 1.5 and 10.0 for NUCLEODUR® 100-5 C_{18} ec.

pH stability of NUCLEODUR® C_{18} ec

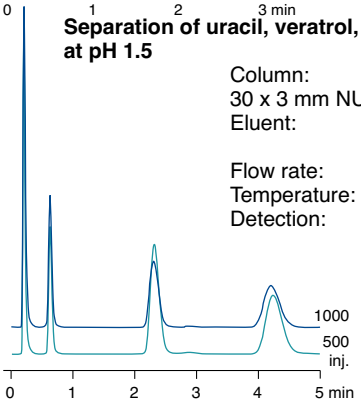
Separation of theophylline and caffeine at pH 10

Column: 30 x 3 mm NUCLEODUR® 100-5 C_{18} ec
Eluent: methanol – aq. NH_3 (20:80, v/v), pH 10
Flow rate: 0.5 ml/min
Temperature: 25 °C
Detection: UV, 254 nm



Separation of uracil, veratrol, toluene and ethylbenzene at pH 1.5

Column: 30 x 3 mm NUCLEODUR® 100-5 C_{18} ec
Eluent: acetonitrile – H_2O (65:35, v/v), TFA, pH 1.5
Flow rate: 1.0 ml/min
Temperature: 25 °C
Detection: UV, 254 nm



NUCLEODUR® octyl phases

In addition to the program of NUCLEODUR® C_{18} phases MACHEREY-NAGEL offers the corresponding octyl modified NUCLEODUR® C_8 Gravity and NUCLEODUR® C_8 ec columns to expand the reversed phase tool box effectively. Based on the same totally spherical and highly pure silica the C_8 phases exhibit the same excellent chemical and mechanical stability features as the C_{18} counterparts. Indeed NUCLEODUR® C_8 Gravity can also be run at pH extremes (pH 1 – 11) by choosing appropriate elution parameters. Due to the shorter chain and less hydrophobic properties of the stationary phase the retention of non-polar compounds is decreased, and in consequence a reduction in time of analysis can be achieved. Moreover a stronger polar selectivity, particularly with the separation of ionizable analytes is frequently observed (as distinct from the C_{18} phases). NUCLEODUR® C_8 ec and NUCLEODUR® C_8 Gravity are most suitable for the development of new methods but also for robust routine analysis.

C_{18} or C_8 · the best of both worlds

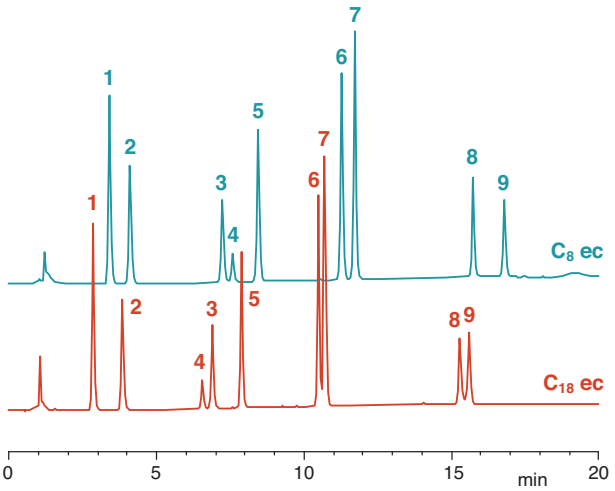
Chromatographers might wonder about the differences between C_8 and C_{18} phases and the preferred range of application. Indeed there are no general guidelines which could make the choice easier but it will always be beneficial to add both phases to the existing pool of reversed phase columns in the laboratory.

However, comparative studies reveal some different selectivity patterns of NUCLEODUR® C_8 ec and NUCLEODUR® C_{18} ec. The separation of phenols on the right shows baseline separation for 2-ethoxyphenol and dimethoxybenzene (veratrol) and in addition a reversal of the elution order of phenol and 4-methoxyphenol can be shown on the octyl phase.

Separation of phenols

Column: 250 x 4 mm NUCLEODUR® 100-5 C_8 ec / C_{18} ec
Eluent: A) water
B) methanol
Gradient for C_8: 2 min 20% B, then to 60% B in 12 min
Gradient for C_{18}: 2 min 25% B, then to 65% B in 12 min
Flow rate: 1.0 ml/min
Temperature: 25 °C
Detection: UV, 275 nm
Injection volume: 10 µl

Peaks:
1. Resorcinol
2. Pyrocatechol
3. 4-Methoxyphenol
4. Phenol
5. 2-Methoxyphenol
6. 2-Ethoxyphenol
7. Veratrol
8. Biphenyl-2-ol
9. Phenetole



MN Appl. No. 120890/120891

C_{18} or C_8 · some general principles

- High density C_8 and C_{18} phases allow tailing-free elution, also for very polar compounds.
- Octyl phases (C_8) show superior polar selectivity.
- Octadecyl phases (C_{18}) show superior hydrophobic selectivity.
- Hydrophobic compounds show shorter retention times on C_8 phases.

NUCLEODUR® C_{18} ec bulk material in 10, 12, 16, 20, 30 and 50 µm for self-packing of prep columns see page 172.

Ordering information

eluent in column acetonitrile / water

Length →	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® 100-3 C_{18} ec	octadecyl phase, particle size 3 µm, 17,5 % C						
Microbore columns							
1 mm ID			717710.10	717711.10	717712.10	717713.10	

Columns with NUCLEODUR® phases

	Length →	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
EC columns								
	2 mm ID	760050.20			760051.20		760052.20	761005.30
	3 mm ID	760050.30			760051.30		760052.30	761005.30
	4 mm ID	760050.40			760051.40		760052.40	761005.40
	4.6 mm ID	760050.46	760046.46	760054.46	760051.46	760053.46	760052.46	761005.40
NUCLEODUR® 100-5 C_{18} ec		octadecyl phase, particle size 5 µm, 17.5 % C						
Microbore columns								
	1 mm ID			717701.10	717700.10	717702.10	717703.10	
EC columns								
	2 mm ID	760004.20			760001.20		760002.20	761100.30
	3 mm ID	760004.30			760001.30		760002.30	761100.30
	4 mm ID	760004.40			760001.40		760002.40	761100.40
	4.6 mm ID	760004.46	760035.46	760013.46	760001.46	760008.46	760002.46	761100.40
VarioPrep columns								
	10 mm ID	762003.100			762029.100		762022.100	762090.80
	21 mm ID	762003.210			762029.210		762022.210	762091.160
	32 mm ID						762022.320	762311.320
	40 mm ID					762027.400	762022.400	762311.320
NUCLEODUR® 100-10 C_{18} ec		octadecyl phase, particle size 10 µm, 17.5 % C						
VarioPrep columns								
	10 mm ID	762011.100			762302.100		762010.100	762090.80
	21 mm ID	762011.210			762302.210		762010.210	762091.160
	32 mm ID						762010.320	762311.320
	40 mm ID					762303.400	762010.400	762311.320
	50 mm ID						762010.500	762311.320

NUCLEODUR® 100-3 C_8 ec		octyl phase, particle size 3 µm, 10.5 % C						
EC columns								
	2 mm ID	760063.20			760060.20		760062.20	761012.30
	3 mm ID	760063.30			760060.30		760062.30	761012.30
	4 mm ID	760063.40			760060.40		760062.40	761012.40
	4.6 mm ID	760063.46	760064.46	760059.46	760060.46	760061.46	760062.46	761012.40
NUCLEODUR® 100-5 C_8 ec		octyl phase, particle size 5 µm, 10.5 % C						
EC columns								
	2 mm ID	760700.20			760701.20		760703.20	761704.30
	3 mm ID	760700.30			760701.30		760703.30	761704.30
	4 mm ID	760700.40			760701.40		760703.40	761704.40
	4.6 mm ID	760700.46	760706.46	760704.46	760701.46	760702.46	760703.46	761704.40
VarioPrep columns								
	10 mm ID	762072.100			762061.100		762062.100	762092.80
	21 mm ID	762072.210			762061.210		762062.210	762093.160
	32 mm ID						762062.320	762321.320
	40 mm ID					762079.400	762062.400	762321.320

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, analytical and preparative main columns in packs of 1.
Other Microbore and VarioPrep columns on request; for available dimensions see page 168/169.

VP guard column cartridge	pack of	for VP column	required holder (see page 168)
VP 10/8, 10 x 8 mm ID	2	10 mm ID	8 mm (REF 718251)
VP 20/16, 20 x 16 mm ID	2	21 mm ID	16 mm (REF 718250)
VP 15/32, 15 x 32 mm ID	1	32 to 50 mm ID	32 mm (REF 718253)

SiOH — unmodified NUCLEODUR® silica for normal phase separations

- **key features:**
  - totally spherical high purity silica
  - pressure stable up to 800 bar
  - suitable for analytical and preparative separation of polar and midpolar compounds
- **technical characteristics:**
  unmodified high purity silica; pore size 110 Å; particle sizes 3 to 50 µm
  pore volume 0.9 ml/g, surface area (BET) 340 m^2/g; pH stability 2 – 8; metal content < 10 ppm (see page 100)
- **recommended application:**
  polar and midpolar compounds under normal phase conditions
  USP L3

Ordering information

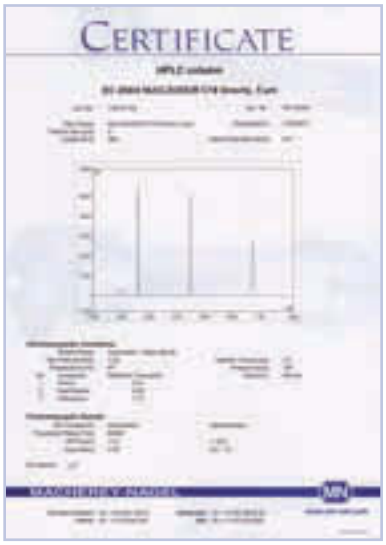
eluent in column *n*-heptane

Length →	50 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® 100-3					particle size 3 µm
EC columns					
4.6 mm ID	**760170.46**		**760172.46**	**760173.46**	**761007.40**
NUCLEODUR® 100-5					particle size 5 µm
EC columns					
4 mm ID				**760007.40**	**761055.40**
4.6 mm ID	**760023.46**		**760012.46**	**760007.46**	**761055.40**
VarioPrep columns					
10 mm ID	**762077.100**	**762078.100**		**762007.100**	**762094.80**
21 mm ID	**762077.210**	**762078.210**		**762007.210**	**762095.160**
32 mm ID				**762007.320**	**762330.320**
40 mm ID			**762075.400**	**762007.400**	**762330.320**

Unmodified NUCLEODUR® bulk material in 10, 12, 16, 20, 30 and 50 µm for self-packing of prep columns see page 172.

Our HPLC QC policy

- **highest production standard**
  our facilities are EN ISO 9001:2008 certified
- **strict quality specifications** for outstanding reliability
- **perfect reproducibility** within each batch and from lot to lot
- Each column is individually tested and supplied with test chromatogram and test conditions

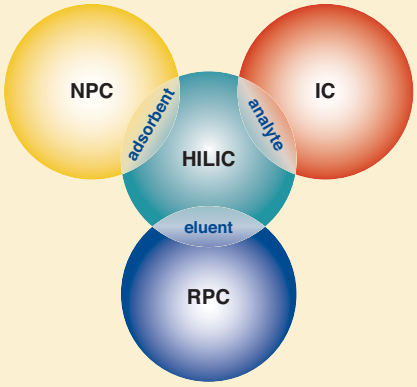


Test mixture for reversed phase columns

Designation	Pack of	REF
Test mixture for reversed phase columns in acetonitrile	1 ml	**722394**

NUCLEODUR® HILIC

zwitterionic phase



- **key features:**
  - ideal for reproducible and stable chromatography of highly polar analytes
  - suitable for analytical and preparative applications as well as **LC–MS**
  - very short column conditioning period
- **technical characteristics:**
  ammonium – sulfonic acid modified silica; pore size 110 Å; particle sizes 1,8, 3 and 5 µm; carbon content 7%; pH stability 2 – 8.5
- **recommended application:**
  hydrophilic compounds such as organic polar acids and bases, polar natural compounds, nucleosides, oligonucleotides, amino acids, peptides, water soluble vitamins

NUCLEODUR® HILIC

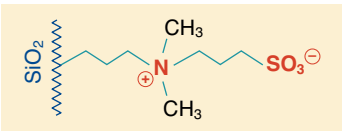
Separation science is always looking for new and effective strategies to accomplish the tasks of modern analytics. Especially for polar compounds reversed phase HPLC – the most common analytical method – is often limited. Here, hydrophilic stationary phases provide an additional tool for the separation of polar analytes in HPLC.

The expression HILIC (Hydrophilic Interaction LIquid Chromatography) was firstly published by Andrew Alpert in 1990 – since then it took quite some efforts to develop robust and reproducible hydrophilic HPLC phases for HILIC chromatography [A. Alpert, J. Chromatography 499 (1990), 177–196].

HILIC combines the characteristics of the 3 major methods in liquid chromatography – reversed phase (RPC), normal phase (NPC) and ion chromatography (IC):

- stationary phases (adsorbents) are mostly polar modifications of silica or polymers (SiOH, NH_2, Diol, (zwitter) ions, ...) – like in NPC
- mobile phases (eluents) are mixtures of aqueous buffer systems and organic modifier like acetonitrile or methanol – like in RPC
- fields of application include quite polar compounds as well as organic and inorganic ions – like in IC

"HILIC is NP chromatography of polar and ionic compounds under RP conditions."



NUCLEODUR® HILIC is a special zwitterionic modified stationary phase based on ultra spherical NUCLEODUR® particles. The betaine character of the ammonium–sulfonic acid ligands results in total charge equalisation and in an overall neutrally charged but highly polar surface.

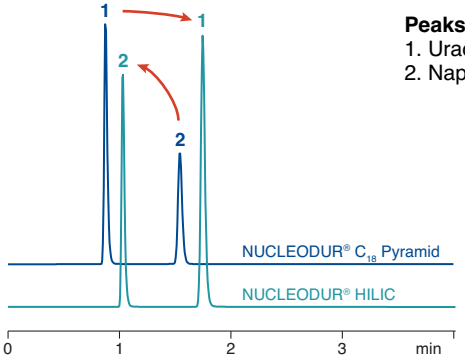
Retention characteristic

Commonly HILIC is described as partition chromatography or liquid/liquid extraction system between the mobile and stationary phase. Versus a water-poor mobile phase a water-rich layer on the surface of the polar stationary phase is formed. Thus, a distribution of the analytes between these two layers will occur.

Furthermore HILIC includes weak electrostatic mechanisms as well as hydrogen donor interactions between neutral polar molecules under high organic elution conditions. This distinguishes HILIC from ion exchange chromatography – main principle for HILIC separation is based on compound's polarity and degree of solvation. More polar compounds will have stronger interaction with the stationary aqueous layer than less polar compounds – resulting in a stronger retention.

Separation of uracil and naphthalene

Columns: 125 x 4 mm NUCLEODUR® C_{18} Pyramid, 3 µm
125 x 4 mm NUCLEODUR® HILIC, 3 µm
Eluent: acetonitrile – water (90:10, v/v)
Flow rate: 1.0 ml/min, temperature 25 °C
Detection: UV, 254 nm



Peaks:
1. Uracil
2. Naphthalene

MN Appl. No. 122911/122912

Nonpolar compounds exhibit faster elution profiles due to minor hydrophobic interactions. Thus, as shown for the separation of uracil and naphthalene the elution order is quite often inverse on HILIC columns compared to RP columns.

In comparison with medium polar aminopropyl phases or modification with less balanced charge equalisation NUCLEODUR® HILIC shows a superb separation and peak shape for critical compounds like adenosine phosphates.

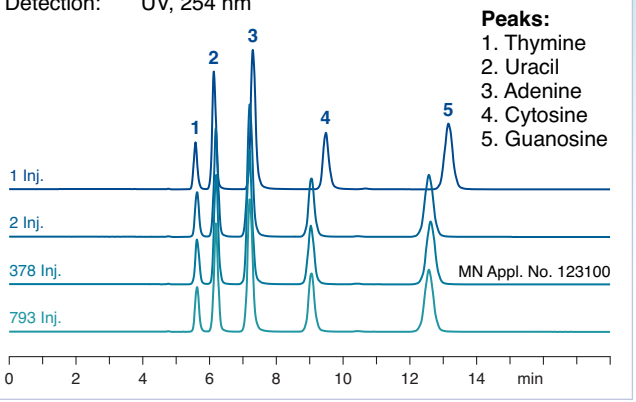
Stability features

Due to an advanced and unique surface modification procedure (pat. pend.) NUCLEODUR® HILIC columns provide short equilibration times – after just 20 min equilibration already the 2nd injection shows stable and reproducible results. Beyond this, NUCLEODUR® HILIC columns are characterized by an outstanding column life time – even after nearly 800 runs the columns show no loss of pristine performance – peak shape and retention are still immaculate.

Due to its high loadability NUCLEODUR® HILIC is absolutely suitable for preparative and semi-preparative applications.

Stability and equilibration

Column: 250 x 4 mm NUCLEODUR® HILIC, 5 µm
Eluent: acetonitrile – 5 mM ammonium acetate (80:20, v/v)
Flow rate: 0.6 ml/min
Temperature: 25 °C
Detection: UV, 254 nm



Overall NUCLEODUR® HILIC provides excellent chromatographic features and is hereby the perfect choice for separation of polar or charged compounds.

Ordering information

eluent in column acetonitrile – water 80:20, v/v

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® HILIC, 1.8 µm								particle size 1.8 µm
EC columns								
2 mm ID	**760521.20**	**760523.20**	**760525.20**	**760526.20**		**760528.20**		
3 mm ID	**760521.30**	**760523.30**						
4 mm ID	**760521.40**	**760523.40**						
4.6 mm ID	**760521.46**	**760523.46**						
NUCLEODUR® HILIC, 3 µm								particle size 3 µm
EC columns								
2 mm ID		**760532.20**			**760531.20**		**760530.20**	**761580.30**
3 mm ID		**760532.30**			**760531.30**		**760530.30**	**761580.30**
4 mm ID		**760532.40**			**760531.40**		**760530.40**	**761580.40**
4.6 mm ID		**760532.46**		**760534.46**	**760531.46**	**760533.46**	**760530.46**	**761580.40**
NUCLEODUR® HILIC, 5 µm								particle size 5 µm
EC columns								
2 mm ID		**760552.20**			**760551.20**		**760550.20**	**761590.30**
3 mm ID		**760552.30**			**760551.30**		**760550.30**	**761590.30**
4 mm ID		**760552.40**			**760551.40**		**760550.40**	**761590.40**
4.6 mm ID		**760552.46**		**760554.46**	**760551.46**	**760553.46**	**760550.46**	**761590.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.
Microbore and VarioPrep columns with NUCLEODUR® HILIC on request; for available dimensions see page 168/169.

NUCLEODUR® CN / CN-RP — cyano-modified high purity silica phase

- **key features:**
  - high retention capacity especially for very polar and unsaturated compounds
  - multi-mode column (RP and NP) widens scope of selectivity
  - stable against hydrolysis at low pH (working range pH 1 – 8)
- **technical characteristics:**
  cyanopropyl-modified high purity silica; pore size 110 Å; particle sizes 3 µm and 5 µm; 7% C; special endcapping
  high reproducibility from lot to lot;
  different retention characteristics in comparison to C_8 and C_{18}
- **recommended application:**
  tricyclic antidepressants
  steroids
  organic acids
  USP L10

Alternative bonded-phase functionality

In reversed phase HPLC it is fairly common to start with C_{18} or C_8 columns, if new methods have to be developed. However, superior polarity and selectivity properties often required for more sophisticated separations, are not always sufficiently provided by classical RP phases, which are usually characterized by a hydrophobic layer of monomeric or polymeric bonded alkylsilanes.

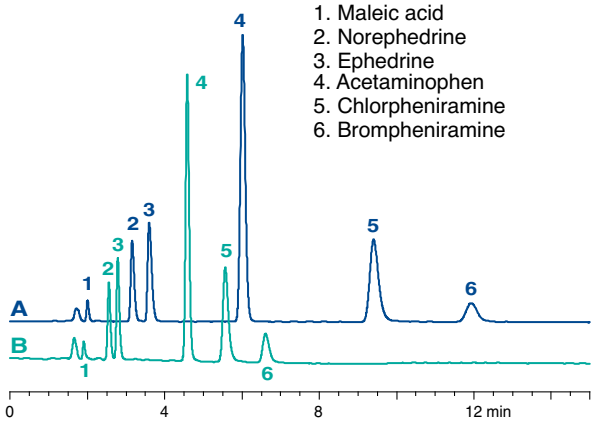
One approach to improve the resolution of compounds poorly separated on nonpolar stationary phases, is to change bonded-phase functionality. The fully end-capped and highly reproducible NUCLEODUR® 100-5 CN-RP phase has cyanopropyl groups on the surface able to generate a clearly recognizable different retention behaviour compared to purely alkyl-functionalized surface modifications (see figure below).

The polarity of the NUCLEODUR® 100-5 CN-RP phase can be classified as intermediate based on multiple retention mechanisms such as dipole-dipole, π-π, and also hydrophobic interactions [C. S. Young and R. J. Weigand, LCGC 20 (2002) 464 – 473]. Therefore, this phase shows a distinct selectivity for polar organic compounds as well as for molecules containing π electron systems (e.g. analytes with double bonds, tricyclic antidepressants) [V. R. Meyer, Practical High Performance Liquid Chromatography (John Wiley & Sons, New York, 3rd ed., 1999)]. Short-chain bonded phases are sometimes suspected of revealing shortcomings in stability towards hydrolysis at low pH [J. J. Kirkland, LCGC 14 (1996) 486 – 500]. The following chromatograms show that even after 100 sample injections and four weeks storage at pH 1 (curve 2), neither a considerable shift in retention, nor a visible change in peak symmetry could be noticed (curve 1 = new column).

Separation of cold medicine ingredients on two different NUCLEODUR® phases

Columns: **A) 250 x 4 mm NUCLEODUR® 100-5 C_{18} ec**
B) 250 x 4 mm NUCLEODUR® 100-5 CN-RP
Eluent: acetonitrile – 100 mM sodium citrate pH 2.5 (15:85, v/v)
Flow rate: 1.0 ml/min, temperature 25 °C
Detection: UV, 270 nm, injection volume: 10 µl

Peaks:
1. Maleic acid
2. Norephedrine
3. Ephedrine
4. Acetaminophen
5. Chlorpheniramine
6. Brompheniramine

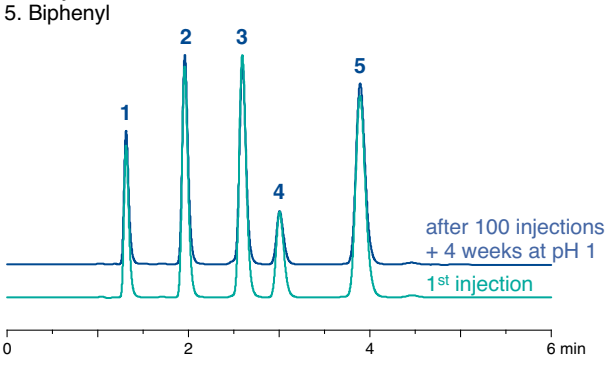


MN Appl. No. 119340

Stability of NUCLEODUR® CN-RP at pH 1

Column: 125 x 4 mm NUCLEODUR® 100-5 CN-RP
Eluent: acetonitrile – water, 2% TFA pH 1 (50:50, v/v)
Flow rate: 1.0 ml/min
Temperature: 25 °C
Detection: UV, 254 nm
Injection volume: 5 µl

Peaks:
1. Benzamide
2. Dimethyl phthalate
3. Phenetole
4. *o*-Xylene
5. Biphenyl



MN Appl. No. 119350

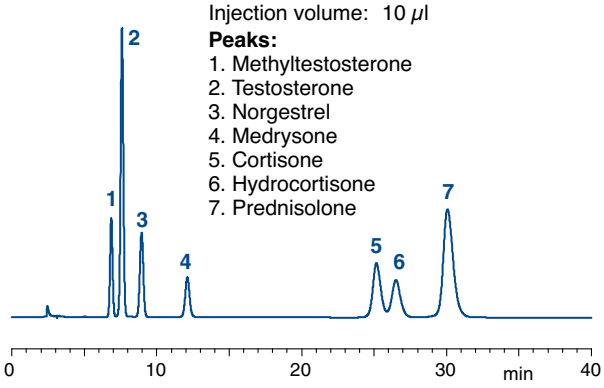
Due to the exceptional polarity features the cyano phase can also be run in the normal phase mode. NUCLEODUR® CN columns for normal phase applications are shipped in *n*-heptane. The drastic change in selectivity and order of elution for a mixture of various steroids in normal and reversed phase mode is displayed in following figure. Moreover the high coverage combined with a thorough endcapping makes NUCLEODUR® 100-5 CN-RP suitable for the separation of ionizable compounds such as basic drugs.

Separation of steroids in normal phase and reversed phase mode

Normal phase mode

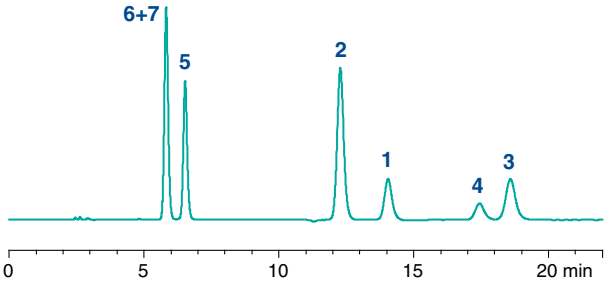
Column:	250 x 4 mm NUCLEODUR® 100-5 CN
Eluent:	*n*-heptane – 2-propanol (90:10, v/v)
Flow rate:	1.0 ml/min
Temperature:	25 °C
Detection:	UV, 254 nm
Injection volume:	10 µl

Peaks:
1. Methyltestosterone
2. Testosterone
3. Norgestrel
4. Medrysone
5. Cortisone
6. Hydrocortisone
7. Prednisolone



Reversed phase mode

Column: 250 x 4 mm NUCLEODUR® 100-5 CN-RP
Eluent: acetonitrile – water (25:75, v/v)
other conditions as for normal phase mode



MN Appl. Nos. 119271/119272

Ordering information

Length →	50 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® 100-3 CN-RP		particle size 3 µm; eluent in column acetonitrile / water			
EC columns					
2 mm ID	**760159.20**	**760157.20**			**761430.30**
3 mm ID		**760157.30**			**761430.30**
4 mm ID			**760156.40**		**761430.40**
4.6 mm ID			**760156.46**		**761430.40**
NUCLEODUR® 100-5 CN-RP		particle size 5 µm; eluent in column acetonitrile / water			
EC columns					
4 mm ID		**760153.40**		**760152.40**	**761420.40**
4.6 mm ID		**760153.46**	**760154.46**	**760152.46**	**761420.40**
NUCLEODUR® 100-5 CN		particle size 5 µm; eluent in column *n*-heptane			
EC columns					
4 mm ID		**760151.40**		**760150.40**	**761419.40**
4.6 mm ID		**760151.46**	**760149.46**	**760150.46**	**761419.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.
Microbore and VarioPrep columns with NUCLEODUR® CN / CN-RP on request; for available dimensions see page 168/169.

NUCLEODUR® NH_2 / NH_2-RP — amino-modified high purity silica

- **key features:**
  - multi-mode columns (for RP, NP and IC)
  - stable against hydrolysis at low pH (working range pH 2 – 8), 100 % stable in water; suitable for **LC/MS**
  - widens scope of analytical HPLC into the polar range
- **technical characteristics:**

  aminopropyl-modified high purity silica; pore size 110 Å; particle sizes 3, 5 and 7 µm; 2.5 % C; not endcapped
- **recommended application:**

  polar compounds under RP conditions (sugars, DNA bases), hydrocarbons under NP conditions

  USP L8

- **normal phase chromatography (NP)** with hexane, dichloromethane or 2-propanol as mobile phase for polar compounds such as substituted anilines, esters, chlorinated pesticides
- **reversed phase chromatography (RP)** of polar compounds in aqueous-organic eluent systems
- **ion exchange chromatography** of anions and organic acids using conventional buffers and organic modifiers

Some compounds, especially polar substances, cannot be sufficiently resolved on C_{18} phases. Polar-modified silica phases offer alternative selectivities such expanding the spectrum of analytical HPLC into the polar range.

Multi-mode columns

Besides cyano modifications, amino modifications belong to the most frequently used polar silica phases – both feature the important advantage, that they can be run in the RP mode using aqueous-organic eluent mixtures as well as in the NP mode e.g. with hexane as mobile phase. NUCLEODUR® Amino, too, belongs to the so-called multi-mode columns.

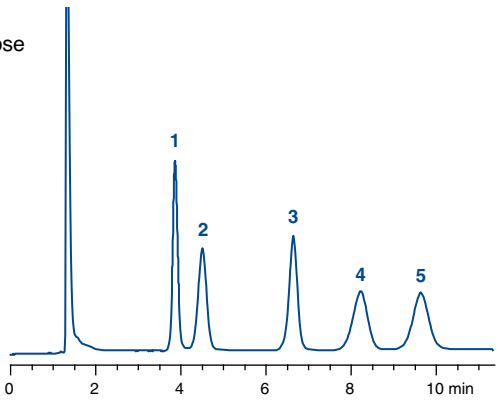
It can be used for reversed phase chromatography (RP) of polar compounds such as sugars in aqueous-organic eluent systems, for normal phase chromatography (NP) of substituted aromatics or chlorinated pesticides with organic mobile phases such as hexane, dichloromethane or 2-propanol, but also for ion exchange chromatography of anions and organic acids using conventional buffers and organic modifiers.

Main field of application of NUCLEODUR® Amino is the separation of simple and complex sugars, sugar alcohols and other hydroxy compounds under RP conditions as well as hydrocarbons under NP conditions.

Reversed phase separation of sugars

Column: 250 x 4 mm NUCLEODUR® 100-5 NH_2-RP
Eluent: acetonitrile – water (79:21, v/v)
Flow rate: 2 ml/min
Detection: RI

Peaks:
1. Fructose
2. Glucose
3. Saccharose
4. Maltose
5. Lactose

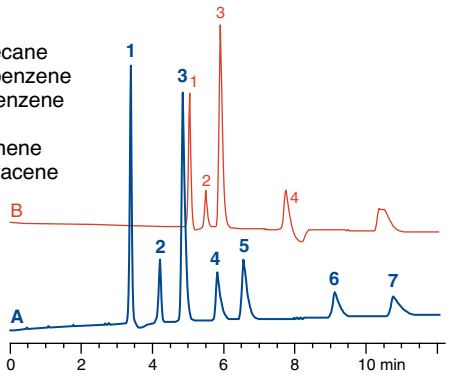


MN Appl. No. 122160

Normal phase separation of middle distillates in accordance with DIN EN 12916

Columns: A) 250 x 4 mm NUCLEODUR® 100-5 NH_2
B) conventional aminopropyl phase
Eluent: heptane
Flow rate: 1 ml/min
Detection: RI

Peaks:
1. Cyclohexane
2. 1-Phenyldodecane
3. 1,2-Dimethylbenzene
4. Hexamethylbenzene
5. Naphthalene
6. Dibenzothiophene
7. 9-Methylanthracene



MN Appl. No. 122180

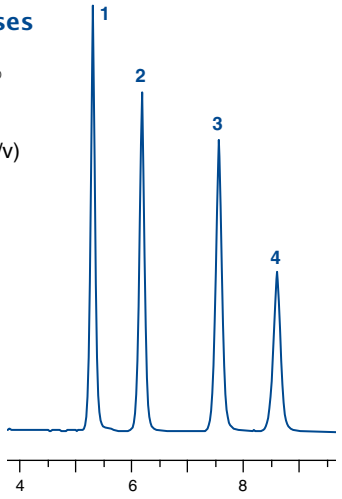
Columns with NUCLEODUR® phases

Even at lower flow rates than for C18 phases, NUCLEODUR® Amino achieves good separations of polar compounds such as DNA bases – this reduces the back pressure as well as the solvent consumption. Even very polar compounds like streptomycin are retained sufficiently for quantitative and qualitative analysis.

Separation of DNA bases

Column: 250 x 4 mm NUCLEODUR® 100-5 NH_2-RP
Eluent: acetonitrile – water (80:20, v/v)
Flow rate: 0.6 ml/min
Temperature: 35 °C
Pressure: 30 bar
Detection: UV, 254 nm

Peaks:
1. Thymine
2. Uracil
3. Cytosine
4. Adenine



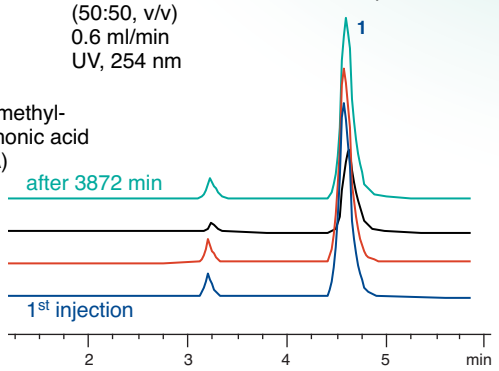
MN Appl. No. 122180

The example below proves the enhanced pH stability of the NUCLEODUR® amino phase and also the outstanding suitability of this column for the separation of total herbicides (AMPA, glyphosate, glufonisate, ...) – you may find the complete application in our online application data base at *www.mn-net.com*.

Hydrolytical resistance for NUCLEODUR® NH_2-RP

Column: 250 x 4 mm NUCLEODUR® 100-5 NH_2-RP
Eluent: acetonitrile – 50 mmol KH_2PO_4, pH 1.75 (50:50, v/v)
Flow rate: 0.6 ml/min
Detection: UV, 254 nm

Peaks:
1. Aminomethyl-phosphonic acid (AMPA)



MN Appl. No. 122190

One of the main problems with conventional amino phases is insufficient resistance towards hydrolysis. Due to a special method of surface modification NUCLEODUR® NH_2 features a pronounced stability at higher as well as at lower pH values. The figure at right shows, that even after several days of exposure of the column material at pH 1.75 good separation efficiency and peak symmetry are maintained. The resulting high column life allows cost reduction due to lower column consumption.

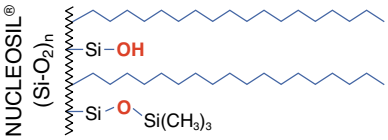
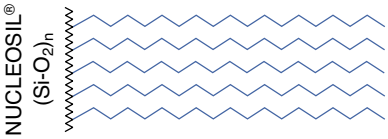
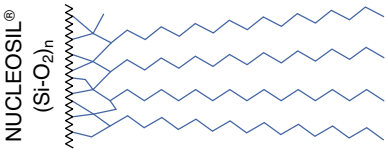
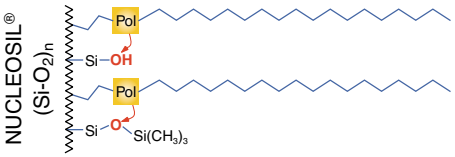
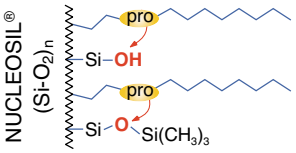
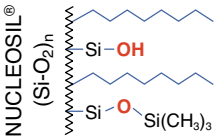
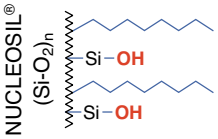
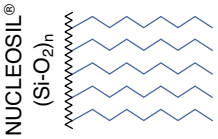
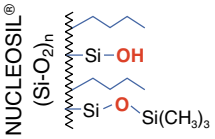
Based on the superspherical silica NUCLEODUR® this phase – like all other members of the NUCLEODUR® family – features a very good pressure stability, which makes it the perfect choice for preparative separations as well as for LC-MS applications. Additionally, the high batch-to-batch reproducibility of NUCLEODUR® NH_2 offers the advantage of reliable analyses especially for routine work.

Ordering information

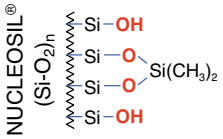
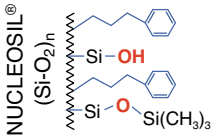
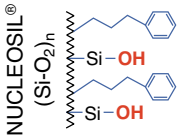
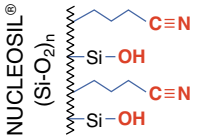
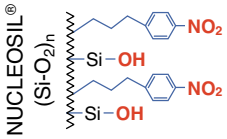
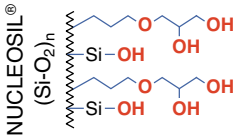
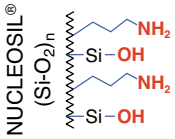
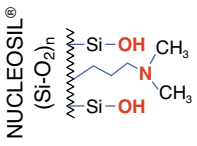
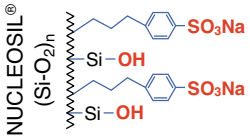
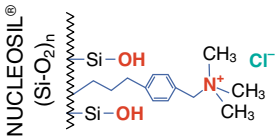
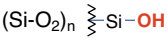
Length →	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEODUR® 100-3 NH_2-RP	particle size 3 µm; eluent in column acetonitrile / water				
EC columns					
2 mm ID	**760740.20**	**760741.20**			**761035.30**
4.6 mm ID			**760742.46**	**760739.46**	**761035.40**
NUCLEODUR® 100-5 NH_2-RP	particle size 5 µm; eluent in column acetonitrile / water				
EC columns					
2 mm ID		**760730.20**		**760732.20**	**761137.30**
3 mm ID		**760730.30**		**760732.30**	**761137.30**
4 mm ID		**760730.40**		**760732.40**	**761137.40**
4.6 mm ID		**760730.46**	**760731.46**	**760732.46**	**761137.40**
NUCLEODUR® 100-5 NH_2	particle size 5 µm; eluent in column *n*-heptane				
EC columns					
4 mm ID		**760720.40**		**760722.40**	**761130.40**
4.6 mm ID		**760720.46**	**760721.46**	**760722.46**	**761130.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.
Microbore and VarioPrep columns with NUCLEODUR® NH_2 / NH_2-RP on request; for available dimensions see page 168/169.

Overview of NUCLEOSIL® HPLC phases

Phase	Specification	Stability	Structure	Separation principle	Page
NUCLEOSIL® RP phases					
C_{18}	octadecyl phase, medium density modification, endcapping 15 % C · USP **L1**	pH 2 – 8		hydrophobic interactions (van der Waals interactions) slight residual silanol interactions	132 – 135
C_{18} HD	octadecyl phase, high density monomeric modification, endcapping 20 % C · USP **L1**	pH 2 – 9		hydrophobic interactions (van der Waals interactions)	134
C_{18} AB	octadecyl phase, special crosslinked modification, endcapping 25 % C · USP **L1**	pH 1 – 9		steric interactions and hydrophobic interactions	134
C_{18} Nautilus	octadecyl phase, embedded polar group, endcapping 16 % C · USP **L60**	pH 2 – 8 up to 100 % H_2O		hydrophobic interactions and polar interactions	134
PROTECT I	special RP phase, protective polar group, monomeric modification, endcapping 11 % C	pH 2 – 8		hydrophobic interactions and polar interactions	135
C_8 ec	octyl phase, medium density modification, endcapping 9 % C · USP **L7**	pH 2 – 8		hydrophobic interactions (van der Waals interactions) slight residual silanol interactions	136
C_8	octyl phase, no endcapping 8.5 % C · USP **L7**	pH 2 – 8		hydrophobic interactions (van der Waals interactions) noticeable silanol interactions	136 – 137
C_8 HD	octyl phase, high density monomeric modification, endcapping 13 % C · USP **L7**	pH 2 – 8		hydrophobic interactions (van der Waals interactions)	137
C_4	butyl phase, medium density modification, endcapping ~ 2 % C · USP **L26**	pH 2 – 8		hydrophobic interactions (van der Waals interactions) residual silanol interactions	137

Widest choice of modifications

Phase	Specification	Stability	Structure	Separation principle	Page
C_2	dimethyl phase 3.5 % C · USP **L16**	pH 2 – 8		hydrophobic interactions (van der Waals interactions) noticeable silanol interactions	138
C_6H_5 ec	phenyl phase, medium density modification, endcapping 8 % C · USP **L11**	pH 2 – 8		π–π interactions and hydrophobic interactions slight residual silanol interactions	174*
C_6H_5	phenyl phase, no endcapping 8 % C · USP **L11**	pH 2 – 8		π–π interactions and hydrophobic interactions noticeable silanol interactions	138
Polar NUCLEOSIL® phases and NUCLEOSIL® ion exchangers					
CN / CN–RP	cyano (nitrile) phase USP **L10**	pH 2 – 8		π–π interactions, polar interactions and hydrophobic interactions	139
NO_2	nitrophenyl	pH 2 – 8		π–π interactions, polar interactions and hydrophobic interactions	174*
OH	diol USP **L20**	pH 2 – 8		polar interactions (hydrogen bonds)	140
NH_2 / NH_2–RP	amino USP **L8**	pH 2 – 8		polar interactions, hydrophobic interactions, weak ion exchange interactions	141
$N(CH_3)_2$	dimethylamino	pH 2 – 8		polar interactions, hydrophobic interactions, weak ion exchange interactions	141
SA	sulphonic acid, strongly acidic cation exchanger (SCX) USP **L9**	pH 2 – 8		strong ion exchange interactions	142
SB	quaternary ammonium groups, strongly basic anion exchanger (SAX) USP **L14**	pH 2 – 8		strong ion exchange interactions	142
Unmodified NUCLEOSIL®	spherical silica · USP **L3**	pH 2 – 8		polar interactions	140

* available as bulk packing (custom-packed columns on request)

Columns with NUCLEOSIL® C_{18} phases

NUCLEOSIL® octadecyl phases (C_{18})

$-(CH_2)_{17}-CH_3$

- **NUCLEOSIL® standard octadecyl phases**
  nonpolar phases · USP **L1**
  pH stability at 20 °C: 2 – 8; carbon content depending on pore size (see ordering information)
- **NUCLEOSIL® C_{18} HD**
  nonpolar hydrophobic high density phases, monomeric modification · USP **L1**
  pH stability 2 – 9; carbon content 20 %
  corresponding NUCLEODUR® phases see C_{18} Gravity page 108 – 111
- **NUCLEOSIL® C_{18} AB**
  crosslinked hydrophobic phase, polymeric modification, inert towards acidic and basic substances with high affinity for silica; pH stability 1 – 9; carbon content 25 % · USP **L1**
  distinct steric selectivity
  corresponding NUCLEODUR® phases see C_{18} Isis page 112 – 113
- **NUCLEOSIL® C_{18} Nautilus**
  stable in 100 % aqueous eluents; carbon content 16 % · USP **L60**
  interesting polar selectivity features
  very good base deactivation
- **wide pore octadecyl phases**
- all octadecyl phases are endcapped

Custom-packed columns with different column dimensions are available on request.

Ordering information

eluent in column acetonitrile / water

Length →	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 50-5 C_{18} ec			particle size 5 µm, pore size 50 Å, 14.5 % C		
EC columns					
4.6 mm ID				**720098.46**	**721829.40**
NUCLEOSIL® 100-3 C_{18}			particle size 3 µm, pore size 100 Å, 15 % C		
EC columns					
4 mm ID		**720150.40**		**720133.40**	**721866.40**
4.6 mm ID	**720841.46**	**720150.46**	**720949.46**	**720133.46**	**721866.40**
ChromCart® cartridges					
4 mm ID		**721883.40**			**721866.40**
4.6 mm ID		**721883.46**	**721806.46**	**721865.46**	**721866.40**
NUCLEOSIL® 100-5 C_{18}			particle size 5 µm, pore size 100 Å, 15 % C		
EC columns					
2 mm ID		**720002.20**		**720014.20**	**721602.30**
3 mm ID		**720002.30**		**720014.30**	**721602.30**
4 mm ID	**720141.40**	**720002.40**	**720120.40**	**720014.40**	**721602.40**
4.6 mm ID	**720141.46**	**720002.46**	**720120.46**	**720014.46**	**721602.40**
ChromCart® cartridges					
2 mm ID		**721622.20**			**721602.30**
3 mm ID		**721622.30**			**721602.30**
4 mm ID		**721622.40**		**721662.40**	**721602.40**
4.6 mm ID		**721622.46**	**721642.46**	**721662.46**	**721602.40**

Columns with NUCLEOSIL® C_{18} phases

Length →	100 mm	125 mm	150 mm	250 mm	Guard columns
VarioPrep columns					
10 mm ID				**715340.100**	**715360.80**
21 mm ID				**715340.210**	**715361.160**
NUCLEOSIL® 100-7 C_{18}			particle size 7 µm, pore size 100 Å, 15 % C		
EC columns					
4 mm ID				**720018.40**	
4.6 mm ID		**720951.46**	**720110.46**	**720018.46**	
VarioPrep columns					
8 mm ID				**715332.80**	**715360.80**
10 mm ID				**715332.100**	**715360.80**
16 mm ID				**715332.160**	**715361.160**
21 mm ID				**715332.210**	**715361.160**
NUCLEOSIL® 100-10 C_{18}			particle size 10 µm, pore size 100 Å, 15 % C		
EC columns					
4 mm ID				**720023.40**	
4.6 mm ID		**720701.46**	**720140.46**	**720023.46**	
ChromCart® cartridges					
4 mm ID				**721689.40**	
4.6 mm ID				**721689.46**	
NUCLEOSIL® 120-3 C_{18}			particle size 3 µm, pore size 120 Å, 11 % C		
EC columns					
4 mm ID	**720149.40**	**720040.40**		**720055.40**	**721606.40**
4.6 mm ID	**720149.46**	**720040.46**	**720740.46**	**720055.46**	**721606.40**
ChromCart® cartridges					
4 mm ID		**721626.40**		**721666.40**	**721606.40**
NUCLEOSIL® 120-5 C_{18}			particle size 5 µm, pore size 120 Å, 11 % C		
EC columns					
4 mm ID		**720051.40**		**720041.40**	**721783.40**
4.6 mm ID		**720051.46**	**720730.46**	**720041.46**	**721783.40**
ChromCart® cartridges					
4 mm ID		**721629.40**		**721712.40**	**721783.40**
NUCLEOSIL® 120-7 C_{18}			particle size 7 µm, pore size 120 Å, 11 % C		
EC columns					
4 mm ID				**720042.40**	

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, analytical and preparative main columns in packs of 1.
ChromCart® columns require the CC connecting kit (REF 721690).
Microbore columns and further VarioPrep columns with NUCLEOSIL® packings are available on request. For possible dimensions see page 168/169.

VP guard column cartridge	pack of	for VP column	required holder (see page 168)
VP 10/8, 10 x 8 mm ID	2	8 and 10 mm ID	8 mm (REF 718251)
VP 20/16, 20 x 16 mm ID	2	16 and 21 mm ID	16 mm (REF 718250)

Columns with NUCLEOSIL® C_{18} phases

	Length →	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 120-10 C_{18}			particle size 10 µm, pore size 120 Å, 11 % C			
EC columns						
	4 mm ID				720043.40	
	4.6 mm ID				720043.46	
NUCLEOSIL® 100-3 C_{18} HD			particle size 3 µm, pore size 100 Å, 20 % C			
EC columns						
	4 mm ID		720191.40			721494.40
	4.6 mm ID		720191.46	720193.46		721494.40
NUCLEOSIL® 100-5 C_{18} HD			particle size 5 µm, pore size 100 Å, 20 % C			
EC columns						
	4 mm ID		720296.40		720280.40	721853.40
	4.6 mm ID		720296.46	720294.46	720280.46	721853.40
ChromCart® cartridges						
	4 mm ID		721852.40		721850.40	721853.40
NUCLEOSIL® 100-5 C_{18} AB			particle size 5 µm, pore size 100 Å, 25 % C			
EC columns						
	4 mm ID		720935.40		720936.40	721603.40
	4.6 mm ID		720935.46	720305.46	720936.46	721603.40
ChromCart® cartridges						
	4 mm ID		721623.40		721663.40	721603.40
NUCLEOSIL® 100-3 C_{18} Nautilus			particle size 3 µm, pore size 100 Å, 16 % C			
EC columns						
	4 mm ID		720472.40			721611.40
	4.6 mm ID		720472.46	720471.46		721611.40
NUCLEOSIL® 100-5 C_{18} Nautilus			particle size 5 µm, pore size 100 Å, 16 % C			
EC columns						
	4 mm ID		720430.40		720431.40	721140.40
	4.6 mm ID		720430.46	720432.46	720431.46	721140.40

Wide pore silica packings

Many biologically interesting molecules can not be separated using conventional narrow pore silicas with pore sizes of about 100 Å.

This is why MACHEREY-NAGEL offers a complete line of wide pore packings with pore sizes of 300, 500, 1000 and 4000 Å. These materials can also be used for size exclusion chromatography (SEC).

	Length →	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 300-5 C_{18}			particle size 5 µm, pore size 300 Å, 6.5 % C			
EC columns						
	4 mm ID				720065.40	721608.40
	4.6 mm ID				720065.46	721608.40

Columns with NUCLEOSIL® C_{18} / Protect I

	Length →	100 mm	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 300-7 C_{18}				particle size 7 µm, pore size 300 Å, 6,5 % C		
VarioPrep columns						
	10 mm ID				**715806.100**	**715360.80**
	21 mm ID				**715806.210**	**715361.160**
NUCLEOSIL® 500-7 C_{18}				particle size 7 µm, pore size 500 Å, 2 % C		
EC columns						
	4.6 mm ID				**720074.46**	
NUCLEOSIL® 1000-7 C_{18}				particle size 7 µm, pore size 1000 Å, ~ 1 % C		
EC columns						
	4.6 mm ID				**720077.46**	
NUCLEOSIL® 4000-7 C_{18}				particle size 7 µm, pore size 4000 Å, < 1 % C		
EC columns						
	4.6 mm ID				**720085.46**	

8 mm ChromCart® guard column cartridges in packs of 3, analytical and preparative main columns in packs of 1.
As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
ChromCart® columns require the CC connecting kit (REF 721690).
Microbore columns and VarioPrep columns with NUCLEOSIL® packings are available on request. For possible dimensions see page 168/169.

VP guard column cartridge	pack of	for VP column	required holder (see page 168)
VP 10/8, 10 x 8 mm ID	2	10 mm ID	8 mm (REF 718251)
VP 20/16, 20 x 16 mm ID	2	21 mm ID	16 mm (REF 718250)

NUCLEOSIL® 100 Protect I — special RP phase with protective polar group

- RP phase with pronounced hydrophilic properties, monomeric coating, endcapped
  carbon content 11 % C

Ordering information

Eluent in column is acetonitrile / water

	Length →	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 100-5 Protect I			particle size 5 µm, pore size 100 Å		
EC columns					
	4 mm ID	**720175.40**		**720170.40**	**721154.40**
	4.6 mm ID	**720175.46**	**720174.46**	**720170.46**	**721154.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.

Analytical columns with NUCLEOSIL® C_8 phases

NUCLEOSIL® octyl phases (C_8)

$-(CH_2)_7-CH_3$

- **NUCLEOSIL® standard octyl phases**
  nonpolar phases for RP and ion-pairing chromatography
  endcapped and non-endcapped modifications available
  pH stability at 20 °C: 2 – 8
- **NUCLEOSIL® C_8 HD**
  nonpolar high density phases, monomeric modification, endcapped;
  corresponding NUCLEODUR® phases see C_8 Gravity page 108 – 111
- recommended for separation of moderately to highly polar (water-soluble) compounds
  applications: steroids, nucleosides, cyclodextrins, pharmacological plant constituents
- all phases: USP **L7**

Custom-packed columns with different column dimensions are available on request

Ordering information

eluent in column acetonitrile / water

	Length →	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 50-5 C_8 ec	particle size 5 µm, pore size 50 Å; endcapped, 9 % C				
EC columns					
	4.6 mm ID			**720092.46**	**721834.40**
NUCLEOSIL® 100-5 C_8 ec	particle size 5 µm, pore size 100 Å; endcapped, 9 % C				
EC columns					
	4 mm ID			**720165.40**	**721805.40**
	4,6 mm ID			**720165.46**	**721805.40**
NUCLEOSIL® 100-5 C_8	particle size 5 µm, pore size 100 Å; not endcapped, 8.5 % C				
EC columns					
	4 mm ID	**720001.40**		**720013.40**	**721601.40**
	4.6 mm ID	**720001.46**	**720990.46**	**720013.46**	**721601.40**
NUCLEOSIL® 100-7 C_8	particle size 7 µm, pore size 100 Å; not endcapped, 8.5 % C				
EC columns					
	4 mm ID			**720017.40**	
	4.6 mm ID			**720017.46**	
NUCLEOSIL® 100-10 C_8	particle size 10 µm, pore size 100 Å; not endcapped, 8.5 % C				
EC columns					
	4 mm ID			**720022.40**	
	4.6 mm ID			**720022.46**	
NUCLEOSIL® 120-3 C_8	particle size 3 µm, pore size 120 Å; not endcapped, 6.5 % C				
EC columns					
	4 mm ID	**720071.40**			**721785.40**
	4.6 mm ID	**720071.46**	**720214.46**		**721785.40**

Analytical columns with NUCLEOSIL® C_8 / C_4

	Length →	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 120-5 C_8	particle size 5 µm, pore size 120 Å; not endcapped, 6.5 % C				
EC columns					
	4 mm ID	**720050.40**		**720052.40**	**721787.40**
	4.6 mm ID	**720050.46**	**720735.46**	**720052.46**	**721787.40**
NUCLEOSIL® 300-5 C_8	particle size 5 µm, pore size 300 Å; not endcapped, ~ 3 % C				
EC columns					
	4.6 mm ID			**720062.46**	**721101.40**
NUCLEOSIL® 100-5 C_8 HD	particle size 5 µm, pore size 100 Å, 13 % C				
EC columns					
	4 mm ID	**720195.40**		**720196.40**	**721500.40**
	4.6 mm ID	**720195.46**	**720194.46**	**720196.46**	**721500.40**

NUCLEOSIL® butyl phases (C_4)

$-(CH_2)_3 - CH_3$

- endcapped phases for RP and ion-pairing chromatography · USP **L26**
- pH stability at 20 °C: 2 – 8; carbon content ~ 2 %
- recommended for separation of macromolecules and hydrophobic substances
- retention times are shorter than on C_8 and C_{18} phases

For butyl phases for biochemical separations please refer to page 159.

Ordering information

eluent in column acetonitrile / water

	Length →	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 120-5 C_4	particle size 5 µm, pore size 120 Å				
EC columns					
	4.6 mm ID			**720096.46**	**721889.40**
NUCLEOSIL® 300-5 C_4	particle size 5 µm, pore size 300 Å				
EC columns					
	4 mm ID	**720901.40**		**720059.40**	**721607.40**
	4.6 mm ID		**720220.46**	**720059.46**	**721607.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.
Microbore and VarioPrep columns with NUCLEOSIL® packings are available on request; for possible dimensions see page 168/169.

Analytical columns with NUCLEOSIL® RP phases

NUCLEOSIL® dimethyl phase (C_2)

$-(CH_3)_2$

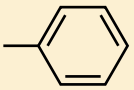
- non-endcapped phase for RP and ion-pairing chromatography · USP **L16**
- pH stability at 20 °C: 2 – 8; carbon content 3.5 %
- retention times are much shorter than for the other RP phases

Ordering information

eluent in column acetonitrile / water

	Length →	250 mm	Guard columns
NUCLEOSIL® 100-7 C_2	particle size 7 µm, pore size 100 Å		
EC columns			
4.6 mm ID		**720089.46**	**721069.40**

NUCLEOSIL® phenyl phases (C_6H_5)

$-(CH_2)_3$ 

- relatively nonpolar, non-endcapped phases for RP and ion pairing chromatography · USP **L11**
- pH stability at 20 °C: 2 – 8; carbon content 8 % C
- polarity similar to C_8, but with different selectivity for polycyclic aromatic hydrocarbons, polar aromatics, fatty acids etc.
- recommended for separation of moderately polar compounds

Ordering information

eluent in column acetonitrile / water

	Length →	250 mm	Guard columns
NUCLEOSIL® 100-5 C_6H_5	particle size 5 µm, pore size 100 Å, not endcapped		
EC columns			
4.6 mm ID		**720956.46**	**721862.40**
NUCLEOSIL® 100-7 C_6H_5	particle size 7 µm, pore size 100 Å, not endcapped		
EC columns			
4 mm ID		**720019.40**	
4.6 mm ID		**720019.46**	

NUCLEOSIL® cyano phases

$-(CH_2)_3-CN$

- polar to mid-polar cyano (nitrile) modified silica · USP **L10**
- for reversed phase and normal phase chromatography:
  normal phase: with low-polarity solvents for many compounds, which can also be separated on unmodified silica, however, due to the rapid equilibration much more suitable for gradient separations
  reversed phase: with different selectivity than C_{18}, C_8 or phenyl modified packings
- pH stability at 20 °C: 2 – 8; carbon content 5 % for 100 Å pores, ~ 3 % for 120 Å pores

Eluent in column is *n*-heptane (except for CN-RP: acetonitrile/water). When using an eluent which is not miscible with *n*-heptane (e.g. water), it is necessary to rinse the column with THF first.

Ordering information

	Length →	250 mm	Guard columns
NUCLEOSIL® 100-5 CN	particle size 5 µm, pore size 100 Å; eluent in column *n*-heptane		
EC columns			
4 mm ID		**720090.40**	**721604.40**
4.6 mm ID		**720090.46**	**721604.40**
NUCLEOSIL® 100-5 CN-RP	particle size 5 µm, pore size 100 Å; eluent in column CH_3CN / H_2O		
EC columns			
4 mm ID		**720205.40**	**721917.40**
4.6 mm ID		**720205.46**	**721917.40**
NUCLEOSIL® 100-10 CN	particle size 10 µm, pore size 100 Å; eluent in column *n*-heptane		
EC columns			
4 mm ID		**720024.40**	
4.6 mm ID		**720024.46**	
NUCLEOSIL® 120-7 CN	particle size 7 µm, pore size 120 Å; eluent in column *n*-heptane		
EC columns			
4 mm ID		**720057.40**	
4.6 mm ID		**720057.46**	

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.

Analytical columns with NUCLEOSIL® OH phases

Unmodified NUCLEOSIL® silica

SiOH

- spherical silica, pH stability 2 – 8 · USP **L3**

Physical properties of unmodified NUCLEOSIL® materials

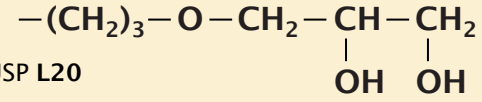
Phase	Pore size	Pore volume	Surface (BET)	Density	Pressure stability
NUCLEOSIL® 50	50 Å	0.8 ml/g	420 m^2/g	0.45 g/ml	600 bar
NUCLEOSIL® 100	100 Å	1.0 ml/g	350 m^2/g	0.36 g/ml	600 bar
NUCLEOSIL® 120	120 Å	0.65 ml/g	200 m^2/g	0.55 g/ml	800 bar
NUCLEOSIL® 300	300 Å	0.8 ml/g	100 m^2/g	0.45 g/ml	400 bar
NUCLEOSIL® 500	500 Å	0.8 ml/g	35 m^2/g	0.45 g/ml	400 bar
NUCLEOSIL® 1000	1000 Å	0.8 ml/g	25 m^2/g	0.45 g/ml	300 bar
NUCLEOSIL® 4000	4000 Å	0.7 ml/g	10 m^2/g	0.48 g/ml	300 bar

Ordering information

Eluent in column is *n*-heptane. When using an eluent which is not miscible with *n*-heptane (e.g. water), it is necessary to rinse the columns with THF first.

	Length →	250 mm	Guard columns
NUCLEOSIL® 50-5		particle size 5 µm, pore size 50 Å	
EC columns			
 4.6 mm ID		**720093.46**	**721600.40**
NUCLEOSIL® 100-5		particle size 5 µm, pore size 100 Å	
EC columns			
 4.6 mm ID		**720099.46**	**721872.40**

NUCLEOSIL® diol phases



- dihydroxypropyl modified silica for RP and NP chromatography · USP **L20**
- less polar than unmodified silica, very easily wettable with water
- pH stability at 20 °C: 2 – 8; carbon content 5 %

Ordering information

Eluent in column is *n*-heptane. When using an eluent which is not miscible with *n*-heptane (e.g. water), it is necessary to rinse the columns with THF first.

	Length →	250 mm	Guard columns
NUCLEOSIL® 100-5 OH (Diol)		particle size 5 µm, pore size 100 Å	
EC columns			
 4.6 mm ID		**720143.46**	**721478.40**
NUCLEOSIL® 100-7 OH (Diol)		particle size 7 µm, pore size 100 Å	
EC columns			
 4.6 mm ID		**720070.46**	

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).

NUCLEOSIL® amino phases

$-(CH_2)_3 - NH_2$

- aminopropyl modified polar silica phase · USP **L8**
- for multi-mode chromatography:

  normal phase chromatography with hexane, dichloromethane or 2-propanol as mobile phase for polar compounds such as substituted anilines, esters, chlorinated pesticides
  reversed phase chromatography of polar compounds like carbohydrates in aqueous-organic eluent systems
  anion exchange chromatography of anions and organic acids using common buffers (e.g. acetate or phosphate) in conjunction with organic modifiers (e.g. acetonitrile)
- pH stability at 20 °C: 2 – 8; carbon content 3.5 %

Eluent in column is *n*-heptane (except for NH_2 RP: acetonitrile/water). When using an eluent which is not miscible with *n*-heptane (e.g. water), it is necessary to rinse the column with THF first.

Ordering information

	Length →	250 mm	Guard columns
NUCLEOSIL® 100-5 NH_2	particle size 5 µm, pore size 100 Å; eluent in column *n*-heptane		
EC columns			
	4.6 mm ID	**720095.46**	**721605.40**
NUCLEOSIL® 100-5 NH_2 RP	particle size 5 µm, pore size 100 Å; eluent in column acetonitrile / water (80:20)		
EC columns			
	4.6 mm ID	**720095.46RP**	**721605.40RP**
NUCLEOSIL® 100-10 NH_2	particle size 10 µm, pore size 100 Å; eluent in column *n*-heptane		
EC columns			
	4.6 mm ID	**720025.46**	

NUCLEOSIL® dimethylamino phase

$-(CH_2)_3 - N(CH_3)_2$

- weakly basic anion exchanger for the separation of many anions
- can also be used in a similar way as the NH_2 phase
- pH stability at 20 °C: 2 – 8; carbon content 4 %

Ordering information

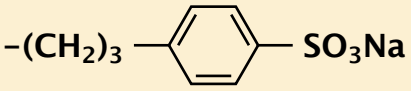
Eluent in column is *n*-heptane. When using an eluent which is not miscible with *n*-heptane (e.g. water), it is necessary to rinse the columns with THF first.

	Length →	250 mm	Guard columns
NUCLEOSIL® 100-5 $N(CH_3)_2$	particle size 5 µm, pore size 100 Å		
EC columns			
	4.6 mm ID	**720994.46**	**721610.40**

8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.

Analytical columns with NUCLEOSIL® SA / SB

NUCLEOSIL® SA phases



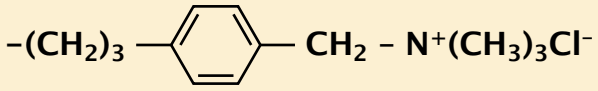
- strongly acidic cation exchangers (SCX) with benzenesulphonic acid modification · USP **L9**
- capacity ~ 1 meq/g
- pH stability at 20 °C: 2 – 8; carbon content 6.5 %

Ordering information

eluent in column 0.15 M $(NH_4)_2HPO_4$, pH 5

Length →	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 100-5 SA			particle size 5 µm, pore size 100 Å	
EC columns				
4 mm ID			720097.40	721487.40
4.6 mm ID	720709.46	720182.46	720097.46	721487.40
ChromCart® cartridges				
4.6 mm ID	721486.46		721342.46	721487.40
NUCLEOSIL® 100-10 SA			particle size 10 µm, pore size 100 Å	
EC columns				
4.6 mm ID			720028.46	721706.40

NUCLEOSIL® SB phases



- strongly basic anion exchangers (SAX) with quaternary ammonium modification · USP **L14**
- capacity ~ 1 meq/g
- pH stability at 20 °C: 2 – 8; carbon content 10 %

Ordering information

eluent in column 0.15 M $(NH_4)_2HPO_4$, pH 5

Length →	125 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 100-5 SB			particle size 5 µm, pore size 100 Å	
EC columns				
4 mm ID			720996.40	721885.40
4.6 mm ID	720989.46	720183.46	720996.46	721885.40
ChromCart® cartridges				
4.6 mm ID	721688.46		721884.46	721885.40
NUCLEOSIL® 100-10 SB			particle size 10 µm, pore size 100 Å	
EC columns				
4.6 mm ID			720029.46	721886.40

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
ChromCart® columns require the CC connecting kit (REF 721690).
8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.

Analytical columns with other RP phases

LiChrospher® · Superspher® packings manufactured by E. Merck (D)

Phase	USP	Particle size	Pore size	Modification	Endcapped	Carbon content
LiChrospher® 100 RP 8, 5 µm	L7	nom. 5 µm	100 Å	octyl	–	12.5%
LiChrospher® 100 RP 8 ec, 5 µm	L7	nom. 5 µm	100 Å	octyl	✔	12.5%
LiChrospher® 100 RP 18, 5 µm	L1	nom. 5 µm	100 Å	octadecyl	–	21%
LiChrospher® 100 RP 18 ec, 5 µm	L1	nom. 5 µm	100 Å	octadecyl	✔	21%
LiChrospher® 60 RP select B, 5 µm	L7	nom. 5 µm	60 Å	octyl	✔	12%
Superspher® 100 RP 18	L1	4 µm	100 Å	octadecyl	–	21%
Superspher® 100 RP 18 ec	L1	4 µm	100 Å	octadecyl	✔	21.6%

all phases as packed ChromCart® cartridges; **eluent in column acetonitrile / water**

Ordering information

Length →	125 mm	150 mm	250 mm	Guard columns
LiChrospher® 100 RP 8, 5 µm				
2 mm ID	728025.20		728026.20	728051.30
3 mm ID	728025.30		728026.30	728051.30
4 mm ID	728025.40		728026.40	728051.40
4.6 mm ID	728025.46	728027.46	728026.46	728051.40
LiChrospher® 100 RP 8 ec, 5 µm				
2 mm ID	728028.20		728029.20	728052.30
3 mm ID	728028.30		728029.30	728052.30
4 mm ID	728028.40		728029.40	728052.40
4.6 mm ID	728028.46	728030.46	728029.46	728052.40
LiChrospher® 100 RP 18, 5 µm				
2 mm ID	728031.20		728032.20	728053.30
3 mm ID	728031.30		728032.30	728053.30
4 mm ID	728031.40		728032.40	728053.40
4.6 mm ID	728031.46	728033.46	728032.46	728053.40
LiChrospher® 100 RP 18 ec, 5 µm				
2 mm ID	728034.20		728035.20	728054.30
3 mm ID	728034.30		728035.30	728054.30
4 mm ID	728034.40		728035.40	728054.40
4.6 mm ID	728034.46	728036.46	728035.46	728054.40
LiChrospher® 60 RP select B, 5 µm				
2 mm ID	728037.20		728038.20	728055.30
3 mm ID	728037.30		728038.30	728055.30
4 mm ID	728037.40		728038.40	728055.40
4.6 mm ID	728037.46	728039.46	728038.46	728055.40
Superspher® 100 RP 18				
2 mm ID	728543.20		728545.20	728546.30
3 mm ID	728543.30		728545.30	728546.30
4 mm ID	728543.40		728545.40	728546.40
4.6 mm ID	728543.46	728544.46	728545.46	728546.40
Superspher® 100 RP 18 ec				
2 mm ID	728540.20		728553.20	728550.30
3 mm ID	728540.30		728553.30	728550.30
4 mm ID	728540.40		728553.40	728550.40
4.6 mm ID	728540.46	728552.46	728553.46	728550.40

ChromCart® columns require the CC connecting kit (REF 721690).
8 mm ChromCart® guard column cartridges in packs of 3, all other columns in packs of 1.

Columns for special HPLC separations

Summary

Anion columns

for analysis of inorganic anions

NUCLEOGEL® Anion I

- strongly basic polymer-based anion exchanger, particle size 10 µm
  pH stability: 1 – 14
- eluent in column 4 mM salicylate buffer pH 7.8
- contrary to the silica-based phase also suited for fluoride analysis

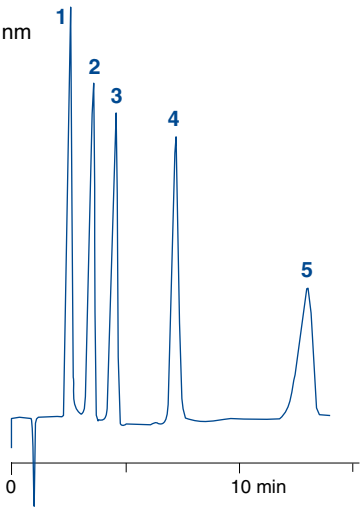
NUCLEOSIL® Anion II

- base material NUCLEOSIL® silica, particle size 10 µm, pore size 300 Å
  strongly basic anion exchanger, exchange capacity 50 µeq/g
  pH stability 2 – 7.5
- eluent in column 2 mM potassium hydrogen phthalate buffer pH 5.6
  recommended buffer concentration for separation of inorganic anions: 2 mmol/l phthalate
- preferred method of detection: conductivity or negative UV detection

Separation of an anion standard

Column: 250 x 4 mm NUCLEOSIL® Anion II
Eluent: 2 mM potassium hydrogen phthalate, pH 5.7
Flow rate: 2 ml/min
Detection: UV, 280 nm

Peaks:
1. $H_2PO_4^-$
2. Cl^-
3. NO_2^-
4. NO_3^-
5. SO_4^{2-}

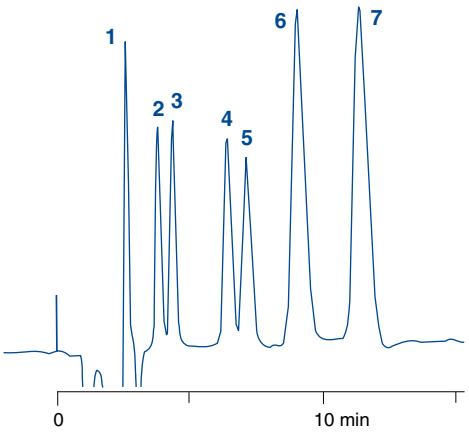


MN Appl. No. 106440

Separation of inorganic anions

Column: 120 x 4.6 mm NUCLEOGEL® Anion I
Eluent: 4 mM salicylic acid / Tris pH 7.8
Flow rate: 1 ml/min
Detection: UV, 254 nm

Peaks:
1. F^-
2. Cl^-
3. NO_2^-
4. Br^-
5. NO_3^-
6. PO_4^{3-}
7. SO_4^{2-}



MN Appl. No. 115050

Ordering information

	Length →	120 mm	250 mm	Guard columns
NUCLEOGEL® Anion I				
Valco type columns				
	4.6 mm ID	**719533**		**719543**
NUCLEOSIL® Anion II				
EC columns				
	4 mm ID		**720094.40**	**721452.40**

NUCLEOGEL® Anion I Valco type guard columns measure 21 x 4 mm and require the guard column holder C (REF 719539, see page 169). Valco type columns in packs of 1, guard columns in packs of 2.
As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359). EC columns and guard column cartridges in packs of 1.

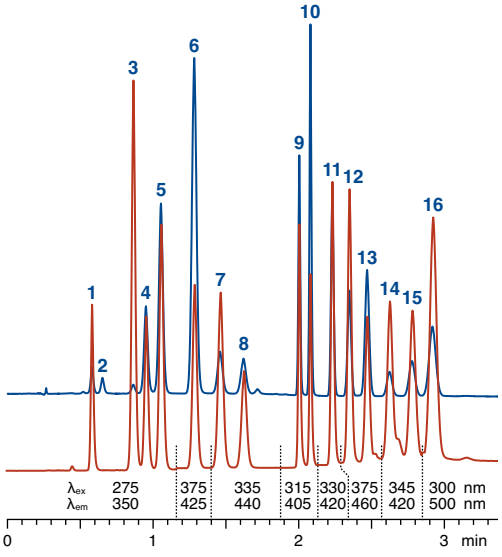
NUCLEODUR® C_{18} PAH — special octadecyl phase for PAH analyses

- base material NUCLEODUR® silica, particle size 3 µm, pore size 110 Å; polymeric coating · USP **L1**
- eluent in column acetonitrile / water 70:30
- allows efficient gradient separation of the 16 PAH according to EPA
- detection of the separated PAH by UV (250 to 280 nm), with diode array or with fluorescence detection at different wavelengths for excitation and emission (acenaphthylene cannot be analysed with fluorescence detection)

Analysis of 16 EPA PAHs with or without acetonitrile

Separation with acetonitrile

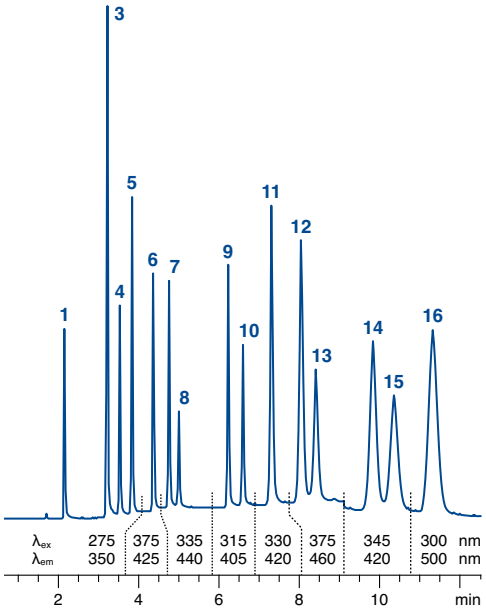
Colum: 100 x 4 mm NUCLEODUR® C_{18} PAH, 3 µm
Eluents: A) methanol – water (80:20, v/v)
B) acetonitrile
Gradient: 2 – 20 % B in 1.2 min, 20 – 100 % B in 0.5 min, 100 % B for 2.5 min, 100 – 2 % B in 0.4 min
Flow rate: 2.5 ml/min
Temperature: 35 °C
Detection: UV, 254 nm
fluorescence (see chromatogram)



MN Appl. No. 123820

Separation without acetonitrile

Colum: 125 x 4 mm NUCLEODUR® C_{18} PAH, 3 µm
Eluents: A) water
B) methanol
Gradient: 65 – 97 % B in 6 min, 97 % B for 5 min, 97 – 65 % B in 0.5 min
Flow rate: 2 ml/min
Temperature: 35 °C
Detection: fluorescence (see chromatogram)



MN Appl. No. 123830

Peaks:

1. Naphthalene
2. Acenaphthylene (not detectable by fluorescence)
3. Acenaphthene
4. Fluorene
5. Phenantrene
6. Anthracene
7. Fluoranthene
8. Pyrene
9. Benz[a]anthracene
10. Chrysene
11. Benzo[b]fluoranthene
12. Benzo[k]fluoranthene
13. Benzo[a]pyrene
14. Dibenz[ah]anthracene
15. Benzo[ghi]perylene
16. Indeno[1,2,3-cd]pyrene

Ordering information

	Length →	100 mm	125 mm	Guard columns
NUCLEODUR® C_{18} PAH, 3 µm				
EC columns				
	3 mm ID	**760783.30**	**760784.30**	**761780.30**
	4 mm ID	**760783.40**	**760784.40**	**761780.40**
PAH standard according to EPA for HPLC				
PAH standard for HPLC	16 PAH according to EPA method 610 in acetonitrile (1 ml) for composition see chromatogram above			**722393**

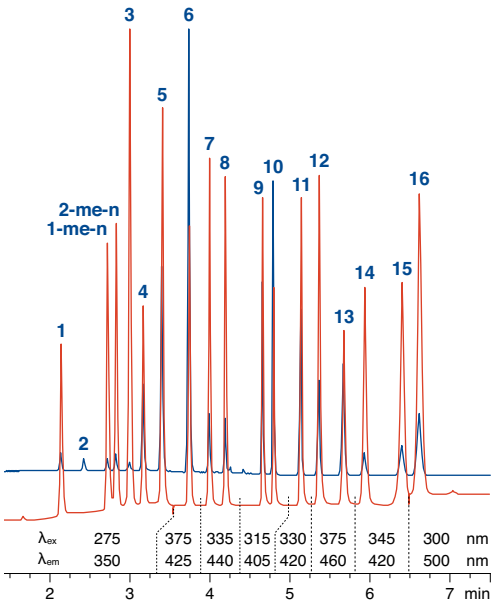
As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, EC columns in packs of 1.

Separation of 18 PAHs on NUCLEODUR® C_{18} PAH

Colum: 125 x 4 mm NUCLEODUR® C_{18} PAH, 3 µm
Eluents: A) methanol – water (70:30, v/v)
B) acetonitrile
Gradient: 0 – 20 % B in 1.5 min,
20 – 50 % B in 1.5 min,
50 – 100 % B in 1.0 min,
100 % B for 3 min,
100 – 0 % B in 0.5 min
Flow rate: 1.5 ml/min
Temperature: 35 °C
Inj. volumes: UV: 1 µl,
fluorescence: 0.5 µl
Detection: UV, 254 nm
fluorescence
(see chromatogram)

Peaks:
(concentrations 10 ng/µl per compound)
1. – 16. see opposite page
1-me-n: 1-methylnaphthalene
2-me-n: 2-methylnaphthalene

MN Appl. No. 123840

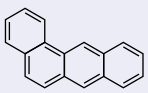


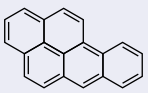
Analysis of polycyclic aromatic hydrocarbons (PAHs) by HPLC

Polycyclic aromatic hydrocarbons (PAHs) are chemical compounds that consist of fused aromatic rings and do not contain heteroatoms or carry substituents. As a pollutant, they are of concern because some compounds have been identified as carcinogenic, mutagenic, and teratogenic. PAHs are natural components of coal or gas. They are delivered to our environment by pyrolysis (incomplete burning) of organic materials like coal, oil, fuel, wood, tabaco, ... and hence can be found globally. Today most PAHs accrue from anthropogenic processes – but also natural origins (forest fire) are possible. Regarding to past pollutions an important impact had production of coke and gas from black coal. Waste products (e.g. tar) from coking or gas plants are often origin of serious ground water pollutions.

Since a number of PAHs (e. g. benzo[a]pyrene, 3-methylcholanthrene and benzanthracene) have been proven to be carcinogenic, control of the PAH content of food, water and soil is an important task for routine analysis. For choice and limiting values of the polycyclics we refer to the governmental regulations, which exist in many countries (e.g. EPA method 610 of the United States Environmental Protection Agency).

PAH can be determined by different chromatographic techniques (TLC, GC, HPLC). Thus the 6 PAHs according to German drinking water specification (TVO) can e.g. be analysed by TLC (see German Standard DIN 38 409), while a much larger number of polycyclic aromatics can be determined by GC or HPLC.


benz[a]anthracene


benzo[a]pyrene

HPLC columns for PAH analysis

For PAH analyses of PAHs we have developed specially modified C_{18} phases based on NUCLEODUR® and NUCLEOSIL® which allow efficient gradient separation of 16 PAHs according to EPA. Detection of the separated PAHs can be achieved by UV (250 – 280 nm), with diode array or with fluorescence detection at different wavelengths for excitation and emission. Acenaphthylene cannot be analysed with fluorescence detection. For cost-effective routine PAH analysis we recommend applications using methanol instead of acetonitrile as eluent. For rapid analysis NUCLEODUR® C_{18} PAH (3 µm) in short columns (100 or 50 mm) provides excellent results at high flow rates. Hereby separation of 16 PAHs according to EPA can be achieved in less than 3 min.

New regulations require determination of 2 additional PAHs (1- and 2-methylnaphthalene) – so we developed highly efficient methods for 18 PAHs on the new NUCLEODUR® C_{18} PAH.

References

Determination of PASH in Diesel fuel by HPLC and photodiode-array detection; J. Bunot, W. Herbel, H. Steinhart, J. High Res. Chrom. **15** (1992) 682 – 685
GIT Spezial Chromat. **2** (1992) 80 – 85

HPLC columns for environmental analyses

NUCLEOSIL® 100-5 C_{18} PAH — special octadecyl phase for PAH analyses

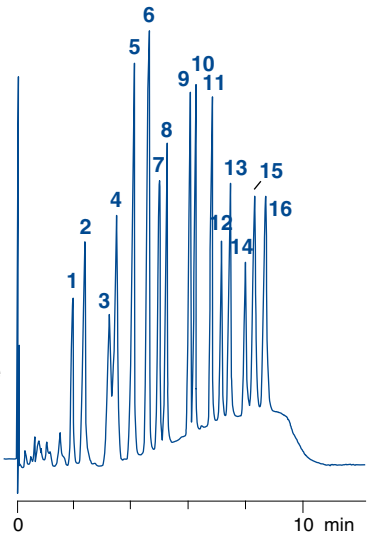
- base material NUCLEOSIL® silica, particle size 5 µm, pore size 100 Å; polymeric coating · USP **L1**
- eluent in column acetonitrile / water 70:30
- allows efficient gradient separation of the 16 PAH according to EPA
- detection of the separated PAH by UV (250 to 280 nm), with diode array or with fluorescence detection at different wavelengths for excitation and emission (acenaphthylene cannot be analysed with fluorescence detection)

Rapid separation of 16 PAH according to EPA

Column:	50 x 4 mm NUCLEOSIL® 100-5 C_{18} PAH
Eluents:	A) water B) acetonitrile
Gradient:	from 55 to 100 % B in 2.5 min; then 3.5 min at 100 % B; finally in 0.1 min from 100 to 55 % B
Flow rate:	1 ml/min
Pressure:	25 – 30 bar
Temperature:	25 °C
Detection:	UV, 260 nm
Injection volume:	10 µl

Peaks:

1. Naphthalene
2. Acenaphthylene
3. Acenaphthene
4. Fluorene
5. Phenantrene
6. Anthracene
7. Fluoranthene
8. Pyrene
9. Benz[a]anthracene
10. Chrysene
11. Benzo[b]fluoranthene
12. Benzo[k]fluoranthene
13. Benzo[a]pyrene
14. Dibenz[ah]anthracene
15. Benzo[ghi]perylene
16. Indeno[1,2,3-cd]pyrene



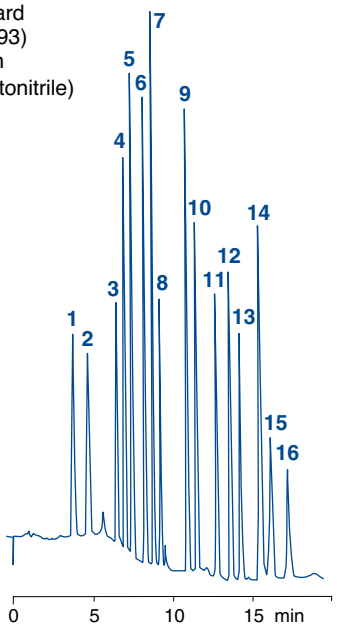
MN Appl. No. 115030

Separation of the PAH standard according to EPA

Column:	150 x 4 mm NUCLEOSIL® 100-5 C_{18} PAH
Eluents:	A) methanol – water (80:20) B) acetonitrile – tetrahydrofuran (93:7)
Gradient:	0 – 100 % B in 10 min, then 5 min at 100 % B
Flow rate:	1 ml/min
Pressure:	140 bar
Temperature:	20 °C
Sample:	PAH standard (REF 722393)
Detection:	UV, 260 nm

Peaks: (10 µg/ml each in acetonitrile)

1. Naphthalene
2. Acenaphthylene
3. Acenaphthene
4. Fluorene
5. Phenantrene
6. Anthracene
7. Fluoranthene
8. Pyrene
9. Benz[a]anthracene
10. Chrysene
11. Benzo[b]fluoranthene
12. Benzo[k]fluoranthene
13. Benzo[a]pyrene
14. Dibenz[ah]anthracene
15. Benzo[ghi]perylene
16. Indeno[1,2,3-cd]pyrene



MN Appl. No. 115040

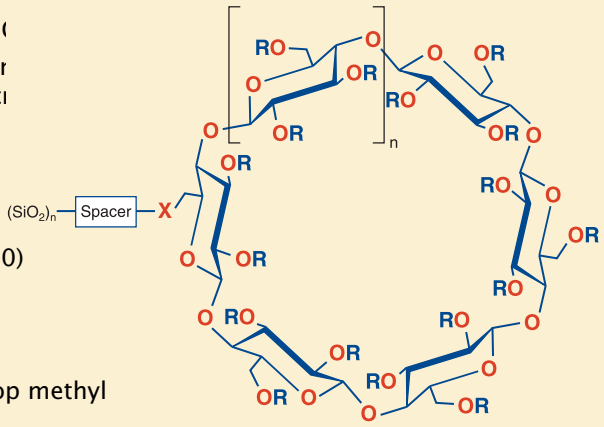
Ordering information

Length →	50 mm	100 mm	150 mm	250 mm	Guard columns
NUCLEOSIL® 100-5 C_{18} PAH					
EC columns					
2 mm ID				720117.20	721599.30
3 mm ID		720819.30	720923.30	720117.30	721599.30
4 mm ID	720756.40	720819.40	720923.40	720117.40	721599.40
4.6 mm ID				720117.46	721599.40
PAH standard according to EPA for HPLC					
PAH standard for HPLC	16 PAH according to EPA method 610 in acetonitrile (1 ml) for composition see chromatogram above				722393

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
8 mm ChromCart® guard column cartridges in packs of 3, EC columns in packs of 1.

NUCLEODEX columns — enantiomer separation based on cyclodextrins

- base material NUCLEOSIL® silica, particle size 5 µm, pore size 100 Å
  modified cyclodextrins as chiral selectors
- **NUCLEODEX β-OH:** β-cyclodextrin (R = H; n = 2) · USP **L45**
  separation based on hydrogen bonds and dipole interactions between functional groups of the analyte and hydroxyl groups of the cyclodextrin
  examples for successful enantiomer separations: chlorthalidone and other compounds, which require free hydroxyl groups for enantioselective interactions
  eluent in column CH_3OH / 0.1% TEAA pH 4 (55:45)
- **NUCLEODEX α-PM:** permethylated α-cyclodextrin (R = (
  for all permethylated phases the ability to form hydroger bonds is reduced, the hydrophobicity of the phase is inci compared to β-OH, resulting in shorter retention times
  examples for successful enantiomer separations: mecoprop and dichlorprop as free carboxylic acids, *trans*-stilbene oxide, styrene oxide
  eluent in column CH_3OH / 50 mM phosphate pH 3 (70:30)



- **NUCLEODEX β-PM:** permethylated β-cyclodextrin (R = CH_3; n = 2) · USP **L45**
  examples for successful enantiomer separations: mephobarbital (prominal), pesticide derivatives mecoprop methyl and dichlorprop methyl
  eluent in column CH_3OH / 0.1% TEAA pH 4 (65:35)
- **NUCLEODEX γ-PM:** permethylated γ-cyclodextrin (R = CH_3; n = 3)
  examples for successful enantiomer separations: steroids or other larger molecules
  eluent in column CH_3OH / 0.1% TEAA pH 4 (55:45)

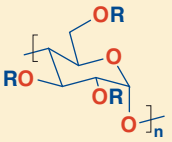
NUCLEODEX phases are especially suited for the control of optical purity, but also for semipreparative separations and for the analysis of positional and *cis-trans* isomers.
For numerous separations on NUCLEODEX phases please visit our website: *www.mn-net.com*.

Ordering information

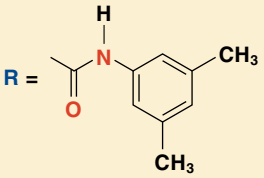
Length →	200 mm	Guard columns
EC columns 		
NUCLEODEX β-OH		
4 mm ID	**720124.40**	**721460.40**
NUCLEODEX α-PM		
4 mm ID	**720127.40**	**721464.40**
NUCLEODEX β-PM		
4 mm ID	**720125.40**	**721462.40**
NUCLEODEX γ-PM		
4 mm ID	**720752.40**	**721466.40**
NUCLEODEX screening kit	**721920**	
consists of one CC 30/4 each with NUCLEODEX β-OH, α-PM, β-PM and γ-PM and a CC column holder 30 mm		

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
All columns and guard column cartridges in packs of 1.

NUCLEOCEL *ALPHA* enantiomer separation based on an amylose derivative



- base material silica,
  chiral selector amylose tris-(3,5-dimethylphenylcarbamate)
  USP **L51**
- similar phases: Chiralpak® AD, Kromasil® AmyCoat™, Europak 01
  high resolution type (S) with 5 µm particle size,
  allows use of shorter columns (150 mm) for faster separations
  pressure stability up to ~150 bar (2000 psi)



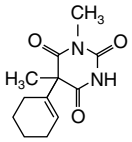
NUCLEOCEL *ALPHA* for normal phase applications:
eluent in column *n*-heptane – propanol-2 (90:10, v/v)
typical eluents are heptane – propanol mixtures

NUCLEOCEL *ALPHA-RP* for reversed phase applications:
eluent in column acetonitrile – water (50:50, v/v)
designed for use either in polar organic mode or with eluents containing high concentrations of chaotropic salts such as perchlorate

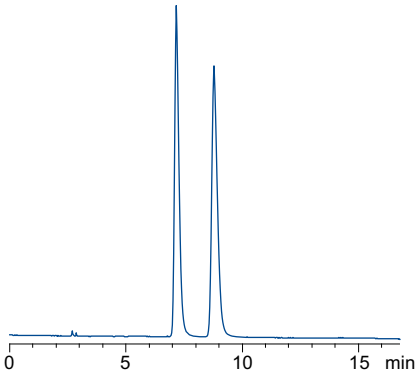
- recommended applications: pharmaceutically active compounds, chiral pollutants (e.g. herbicides, PCB), chiral compounds in food (dyes, preservatives), chiral catalysts and bioorganic compounds

Enantiomer separation of hexobarbital

Column:	250 x 4.6 mm NUCLEOCEL *ALPHA S*
Eluent:	*n*-heptane – 2-propanol (80:20, v/v)
Flow rate:	1 ml/min
Temperature:	22 °C
Detection:	UV, 210 nm
Injection volume:	5 µl
Concentration:	1 µg/µl



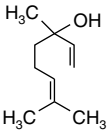
$\alpha = 1.39$
$R_s = 3.78$



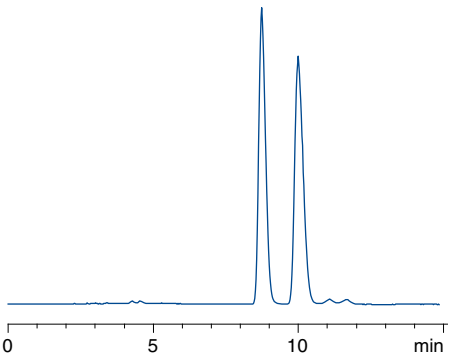
MN Appl. No. 121940

Enantiomer separation of linalool

Column:	250 x 4.6 mm NUCLEOCEL *ALPHA-RP S*
Eluent:	acetonitrile – water (50:50, v/v)
Flow rate:	1 ml/min
Temperature:	35 °C
Detection:	UV, 210 nm
Injection volume:	5 µl
Concentration:	1 µg/µl



$\alpha = 1.21$
$R_s = 2.44$



MN Appl. No. 121920

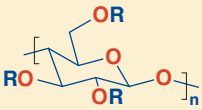
Ordering information

	Length →	150 mm	250 mm	Guard columns
EC columns 	**NUCLEOCEL *ALPHA S*, 5 µm**			
	4.6 mm ID	**720644.46**	**720645.46**	**721000.40**
	NUCLEOCEL *ALPHA-RP S*, 5 µm			
	4.6 mm ID	**720654.46**	**720655.46**	**721001.40**

As guard columns for 4.6 mm EC columns use 4 mm ID ChromCart® guard column cartridges with guard column adaptor EC (REF 721359). All columns and guard columns in packs of 1.

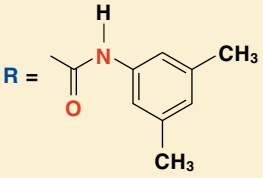
NUCLEOCEL *DELTA* enantiomer separation based on a cellulose derivative

- base material silica,
  chiral selector cellulose tris-(3,5-dimethylphenylcarbamate)
  USP **L40**
- similar phases: Chiralcel® OD, Kromasil® CelluCoat™, Eurocel® 01, Lux™ Cellulose-1



high resolution type (S) with 5 µm particle size,
allows use of shorter columns (150 mm) for faster separations
pressure stability up to ~150 bar (2000 psi)

NUCLEOCEL *DELTA* for normal phase applications:
eluent in column *n*-heptane – propanol-2 (90:10, v/v)
typical eluents are heptane – propanol mixtures

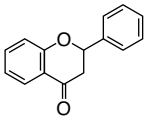


NUCLEOCEL *DELTA-RP* for reversed phase applications:
eluent in column acetonitrile – water (40:60, v/v)
designed for use either in polar organic mode or with eluents containing high concentrations of chaotropic salts such as perchlorate

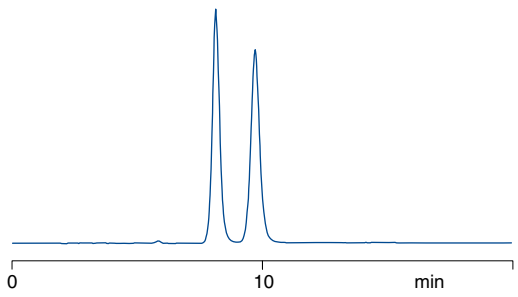
- recommended applications: pharmaceutically active compounds, chiral pollutants (e.g. herbicides, PCB), chiral compounds in food (dyes, preservatives), chiral catalysts and bioorganic compounds

Enantiomer separation of flavanone

Column: 250 x 4.6 mm NUCLEOCEL *DELTA S*
Eluent: *n*-heptane – 2-propanol (90:10, v/v)
Flow rate: 1 ml/min
Temperature: 25 °C
Detection: UV, 254 nm
Injection volume: 5 µl
Concentration: 1 µg/µl



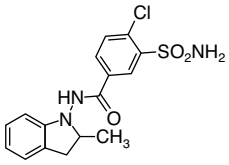
α = 1.29
R_s = 2.6



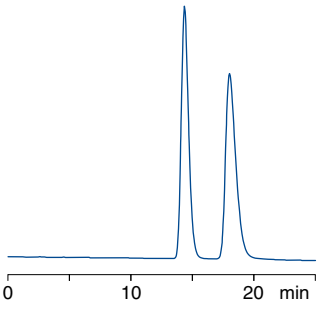
MN Appl. No. 121260

Enantiomer separation of indapamide

Column: 250 x 4.6 mm NUCLEOCEL *DELTA-RP S*
Eluent: acetonitrile – water (40:60, v/v)
Flow rate: 0.5 ml/min
Temperature: 40 °C
Detection: UV, 254 nm
Injection volume: 5 µl
Concentration: 1 µg/µl



α = 1.3
R_s = 2.6



MN Appl. No. 121230

Ordering information

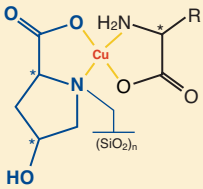
	Length →	150 mm	250 mm	Guard columns
EC columns 	**NUCLEOCEL *DELTA S*, 5 µm**			
	4.6 mm ID	**720446.46**	**720445.46**	**721002.40**
	NUCLEOCEL *DELTA-RP S*, 5 µm			
	4.6 mm ID	**720451.46**	**720450.46**	**721003.40**

As guard columns for 4.6 mm EC columns use 4 mm ID ChromCart® guard column cartridges with guard column adaptor EC (REF 721359). All columns and guard columns in packs of 1.

VarioPrep columns with NUCLEOCEL *ALPHA* and NUCLEOCEL *DELTA* on request; for available dimensions see page 168

HPLC columns for enantiomer separation

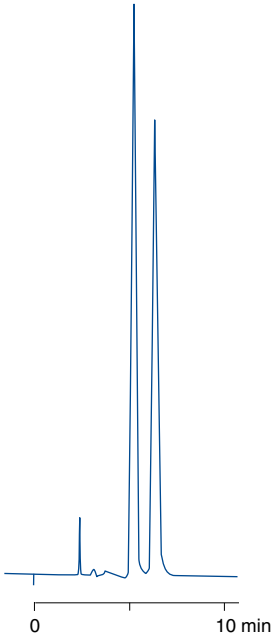
NUCLEOSIL® CHIRAL-1 enantiomer separation based on ligand exchange



- base material NUCLEOSIL® silica, particle size 5 µm, pore size 120 Å
- chiral selector L-hydroxyproline / Cu^{2+} complexes · USP **L32**
- principal interaction mode:
  formation of ternary mixed-ligand complexes with Cu(II) ions
  differences in the stability of the diastereomeric complexes cause chromatographic separation
- eluent in column 0.5 mM copper sulphate solution
- recommended application: enantiomers with two polar functional groups with the correct spacing such as α-amino acids, α-hydroxycarboxylic acids (e.g. lactic acid), *N*-alkyl-α-amino acids etc.

Separation of *D,L*-alanine enantiomers

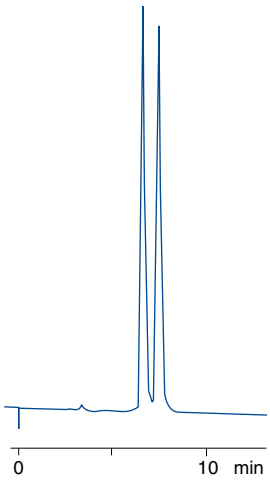
Column:	250 x 4 mm NUCLEOSIL® CHIRAL-1
Eluent:	0.5 mM $CuSO_4$
Flow rate:	1 ml/min
Pressure:	60 bar
Temperature:	60 °C
Detection:	UV, 250 nm



MN Appl. No. 105410

Separation of *D,L*-threonine enantiomers

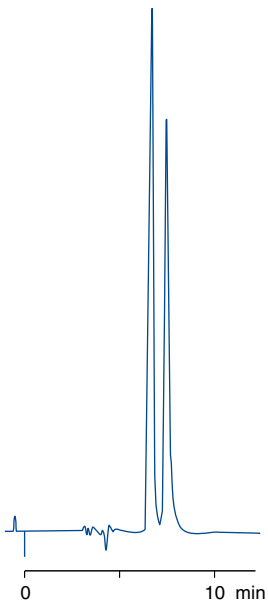
Column:	250 x 4 mm NUCLEOSIL® CHIRAL-1
Eluent:	0.25 mM $CuSO_4$
Flow rate:	0.8 ml/min
Pressure:	65 bar
Temperature:	60 °C
Detection:	UV, 240 nm



MN Appl. No. 105410

Enantiomer separation of lactic acid

Column:	250 x 4 mm NUCLEOSIL® CHIRAL-1
Eluent:	0.5 mM $CuSO_4$
Flow rate:	0.8 ml/min
Temperature:	80 °C
Detection:	UV, 240 nm
Injection volume:	1 µl



MN Appl. No. 105560

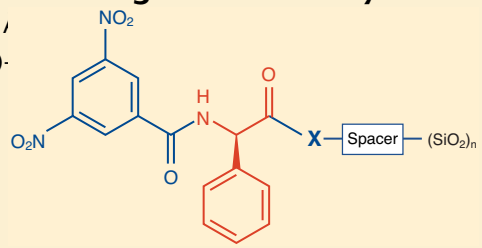
Ordering information

		Length →	250 mm	Guard columns
NUCLEOSIL® CHIRAL-1				
EC columns				
	4 mm ID		**720081.40**	**721455.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
All columns and guard columns in packs of 1.

NUCLEOSIL® CHIRAL-2 / NUCLEOSIL® CHIRAL-3

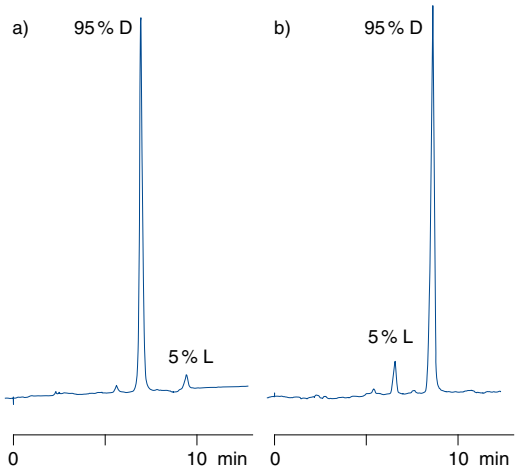
enantiomer separation in organic eluent systems



- base material NUCLEOSIL® silica, particle size 5 µm, pore size 100 Å
- chiral selector for NUCLEOSIL® CHIRAL-2 is *N*-(3,5-dinitrobenzoyl)-*D*-phenylglycine, for CHIRAL-3 the optical antipode is used, "brush type" phases · CHIRAL-3 = USP **L36**
- principle interaction modes: charge-transfer interactions, hydrogen bonds, dipole-dipole interactions and steric effects
- eluent in column *n*-heptane / 2-propanol / TFAA 100:0.5:0.5
- recommended application: analysis of stereoisomers such as separation of enantiomers and diastereomers, control of optical purity of plant protectives (pesticides, e.g. propionic acid derived herbicides) pharmaceuticals etc. and for product control in chiral organic syntheses
- For control of the optical purity of a substance, the two columns NUCLEOSIL® CHIRAL-2 and NUCLEOSIL® CHIRAL-3 allow to select conditions such that the minor enantiomer, which is present as an impurity, is eluted before the main peak. Thus, overlapping peaks are avoided. This makes an exact quantification of the impurity much easier.

Control of optical purity of mecoprop methyl

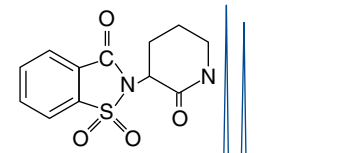
Columns:	250 x 4 mm a) NUCLEOSIL® CHIRAL-2 b) NUCLEOSIL® CHIRAL-3
Eluent:	*n*-heptane – 2-propanol – TFA (100:0.05:0.05, v/v/v)
Flow rate:	1 ml/min
Temperature:	ambient
Detection:	UV, 230 nm
Injection volume:	1 µl (sample with 90 % ee)



MN Appl. No. 111360

Enantiomer separation of *D,L*-supidimide

Column:	250 x 4 mm NUCLEOSIL® CHIRAL-2
Eluent:	tetrahydrofuran – *n*-heptane (10:3, v/v)
Flow rate:	1.0 ml/min
Detection:	UV, 220 nm



MN Appl. No. 105690

Ordering information

		Length →	250 mm	Guard columns
EC columns	**NUCLEOSIL® CHIRAL-2**			
	4 mm ID		**720088.40**	**721458.40**
	NUCLEOSIL® CHIRAL-3			
	4 mm ID		**720350.40**	**721458.40**

8 x 4 mm ID ChromCart® guard column cartridges for NUCLEOSIL® CHIRAL-2 and CHIRAL-3 are identical and used with guard column adaptor EC (REF 721359). They are supplied in packs of 3, the EC columns in packs of 1.

RESOLVOSIL BSA-7 — protein phase for enantiomer separation

- base material NUCLEOSIL® silica, particle size 7 µm, pore size 300 Å
- chiral selector bovine serum albumin (BSA)
- separation based on selective interaction of proteins with low molecular compounds, i.e. principles of bioaffinity, including hydrophobic interactions (similar to a true reversed phase), interactions of polar groups and steric effects
- eluent in column 0.1 M phosphate buffer pH 7.5, 2% 1-propanol
- recommended applications: amino acid derivatives, aromatic amino acids, aromatic sulphoxides, barbiturates, benzodiazepinones, benzoin and benzoin derivatives, β-blockers, coumarin derivatives, and for monitoring stereoselective microbial and enzymatic conversions

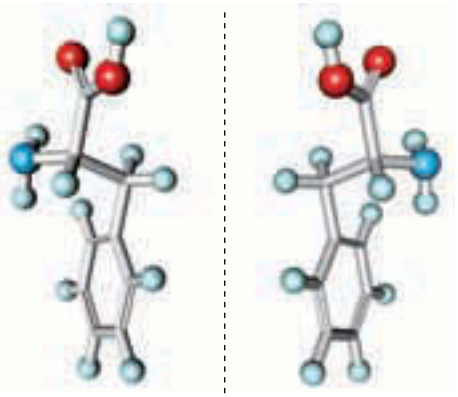
Enantiomer separation of *N*-benzoyl-*D,L*-amino acids

S. Allenmark et al. in "Affinity chromatography and biological recognition" (I. Chaiken, M. Wilchek, and I. Parikh. Eds.), Academic Press, New York, 1983, p. 259 – 260

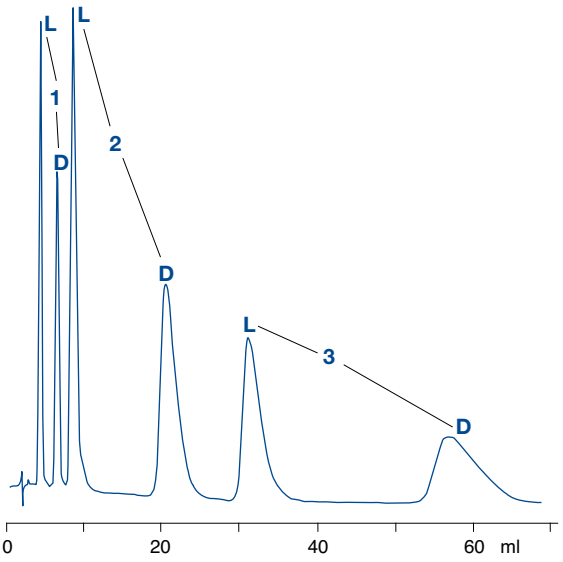
Column: 150 x 4 mm RESOLVOSIL BSA-7
Eluent: 50 mM phosphate buffer pH 6.5 + 1 % 1-propanol
Flow rate: 0.70 ml/min
Detection: UV, 225 nm

Peaks:

1. Serine
2. Alanine
3. Phenylalanine



MN Appl. No. 105450



Ordering information

	Length →	150 mm	Guard column
RESOLVOSIL BSA-7			
EC columns			
	4 mm ID	**720046.40**	**721702.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
EC columns and guard columns in packs of 1.

NUCLEOSIL® 4000-7 PEI — anion exchange of proteins and peptides

- base material NUCLEOSIL® silica, particle size 7 µm, pore size 4000 Å
  polymeric, covalently bonded polyethyleneimine network, weakly basic anion exchanger
  ion exchange capacity 0.15 mmol/g; protein binding capacity 61 mg BSA/g
- pH stability 2 – 8.5; max. working pressure 250 bar
- separation principle: reversible adsorption of negatively charged substances to positively charged groups on the exchanger material and their subsequent displacement by either increasing ionic strength or pH changes in the mobile phase
- high selectivity for numerous proteins; e.g β-lactoglobulins A and B, two proteins differing in just two amino acids, can be separated in only 10 minutes; biological activity of purified proteins is preserved
- good binding and desorption kinetics for nucleotides as well
- eluent in column methanol
- more examples for the purification of different peptides and proteins can be found in our application database at *www.mn-net.com*

Recovery of proteins

Column: 50 x 4 mm NUCLEOSIL® 4000-7 PEI
Eluent: 10 mM NaH_2PO_4, 1.5 M NaCl, pH 7.0
Flow rate: 1 ml/min
Sample: 50 µg of each protein

Protein	Recovery [%]
Myoglobin	100
Transferrin	95
Ovalbumin	98
Bovine serum albumin	100
Glucose oxidase	100
α-Amylase	100
Soybean trypsin inhibitor	100
β-Lactoglobulin	97
Ferritin	85

Recovery of specific enzyme activity after HPLC

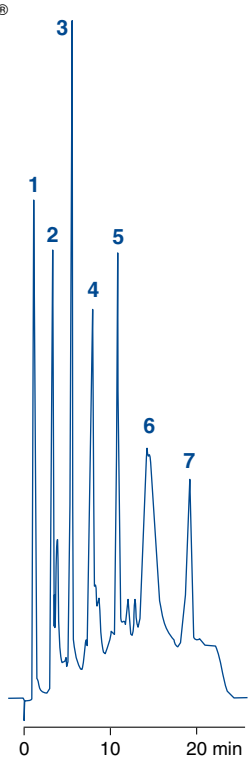
Columns: 50 x 4 mm NUCLEOSIL® 4000-7 PEI
Eluents: A) 20 mM Tris-HCl pH 8.5; B) A + 1.5 M NaCl
Gradient: 0 – 100 % B in 5 min, 1 ml/min, 30 bar
Detection: UV, 280 nm

Enzyme	Recovery [%]
Catalase (bovine liver)	93
L-Lactic dehydrogenase LDH-1 isoenzyme (porcine heart)	102
Callicrein (porcine pancreas)	98
Glucose oxidase (Aspergillus niger)	104
Peroxidase (horseradish)	100

Separation of protein standards

Column: 125 x 4 mm NUCLEOSIL® 4000-7 PEI
Eluents: A) 2 mM Tris / acetate pH 8.0
B) 20 mM Tris / acetate pH 8.0 + 1.5 M KCl
Gradient: linear 0 – 40 % B in 20 min
Flow rate: 1 ml/min
Pressure: 76 bar
Detection: UV, 280 nm
Inj. volume: 20 µl

Peaks:
1. Catalase
2. Myoglobin
3. α-Amylase
4. Transferrin
5. α-Lactalbumin
6. Glucose oxidase
7. Soybean trypsin inhibitor



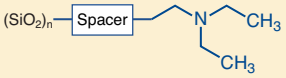
MN Appl. No. 108310

Ordering information

	Length →	125 mm	250 mm	Guard columns
NUCLEOSIL® 4000-7 PEI				
EC analytical columns	4 mm ID	**720402.40**		**721091.40**
VarioPrep prep. columns	10 mm ID	**715230.100**	**715231.100**	

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
Guard columns in packs of 3, other columns in packs of 1.

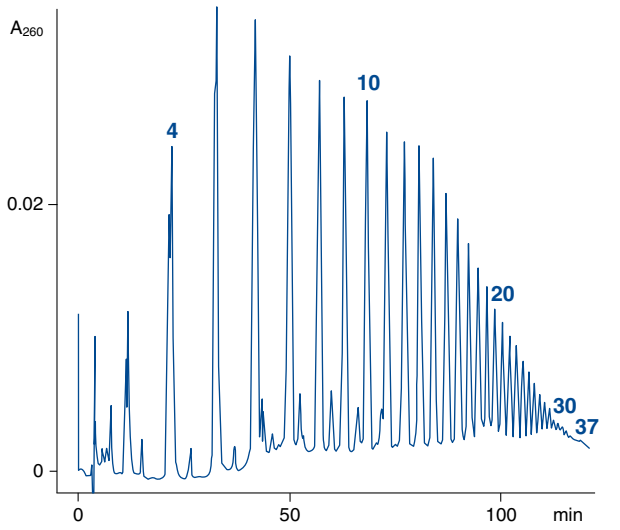
NUCLEOGEN® columns anion exchange chromatography of nucleic acids



- base material silica, particle size 7 µm
  DEAE anion exchanger
- **NUCLEOGEN® 60-7 DEAE:** pore size 60 Å
  for separation of oligonucleotides up to chain lengths of 40 bases with recoveries > 95 %
  capacity 200 A_{260}/ml (~ 300 A_{260} for a 125 x 4 mm ID column, 1875 A_{260} for a 125 x 10 mm ID column); preparative separations possible when using higher flow rates and longer gradient times
- **NUCLEOGEN® 500-7 DEAE:** pore size 500 Å
  for separation of tRNA, 5S RNA, viroids and messenger RNA in the intermediate molecular weight range (25,000 – 1,000,000 daltons) with recoveries > 95 %
  capacity 730 A_{260} for a 125 x 6 mm ID column, 1940 A_{260} for a 125 x 10 mm ID column
- **NUCLEOGEN® 4000-7 DEAE:** pore size 4000 Å
  for separation of plasmids, DNA restriction fragments, ribosomal RNA, messenger RNA and viral RNA, i. e. very high molecular weight nucleic acids (e. g. 1 – 50 megadaltons)
  capacity 120 A_{260} for a 125 x 6 mm ID column, 350 A_{260} for a 125 x 10 mm ID column
- eluent in column methanol
- for more separations of deoxyoligonucleotides, plasmids and DNA restriction fragments visit our website *www.mn-net.com*

Separation of oligo(rA)$_n$

Column:	125 x 4 mm NUCLEOGEN® 60-7 DEAE
Buffers:	A) 20 mM phosphate, pH 5.5, 5 M urea B) buffer A + 1 M KCl
Gradient:	0 – 100 % B in 200 min
Flow rate:	2 ml/min, 110 bar
Temperature:	ambient
Detection:	UV, 260 nm

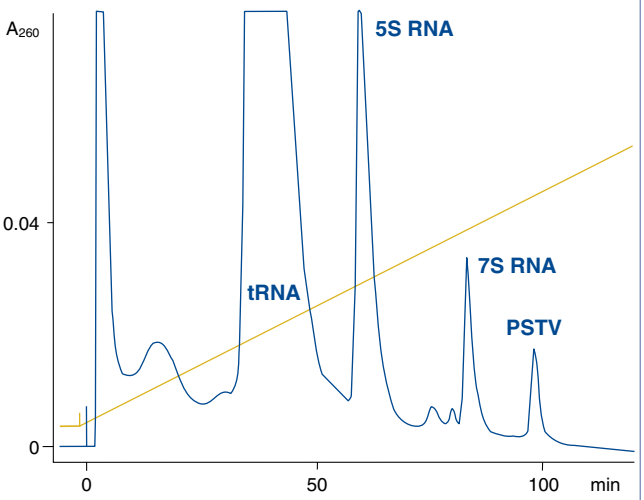


MN Appl. No. 115180

Preparative separation of a crude RNA extract of viroid (PSTV) infected tomato plants

D. Riesner, BioEngineering **1** (1988) 42 – 48

Column:	125 x 6 mm NUCLEOGEN® 500-7 DEAE
Buffers:	A) 250 mM KCl, 20 mM phosphate buffer pH 6.6, 5 M urea B) 1 M KCl, 20 mM phosphate buffer pH 6.6, 5 M urea
Gradient:	0 – 50 % B in 120 min, 50 – 100 % B in 250 min
Flow rate:	3 ml/min, 40 bar
Temperature:	ambient
Detection:	260 nm



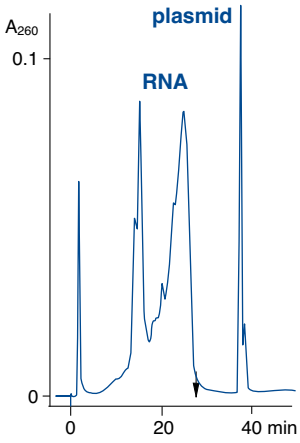
MN Appl. No. 107490

Separation of plasmid pBR 322

M. Colpan, D. Riesner, private communication

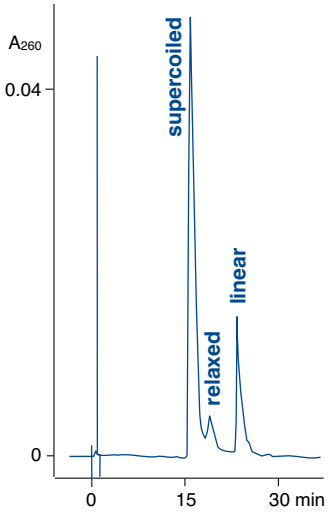
A) isolation of plasmid DNA from a crude cell lysate

Sample: 5 µg plasmid pBR 322 containing cleared lysate from *E. coli*
Column: 125 x 6 mm NUCLEOGEN® 4000-7 DEAE
Eluents: A) 20 mM K phosphate buffer pH 6.9; 5 M urea
B) eluent A + 1.5 M KCl
Gradient: 20 % – 100 % B in 50 min;
arrow = ionic strength of 850 mM
Flow rate: 1.0 ml/min, 70 bar, ambient temperature
Detection: UV, 260 nm



B) separation of supercoiled plasmid from relaxed and linear forms

Sample: plasmid pBR 322, supercoiled, relaxed and linear
Column: 125 x 6 mm NUCLEOGEN® 4000-7 DEAE
Eluents: A) 20 mM phosphate buffer pH 6.8; 6 M urea
B) eluent A + 2 M KCl
Gradient: 42 % – 100 % B in 230 min
Flow rate: 1.5 ml/min, 45 bar, ambient temperature



MN Appl. No. 107480

Ordering information

Length →	125 mm	Guard columns
NUCLEOGEN® 60-7 DEAE		
EC analytical columns		
4 mm ID	**736596.40**	**736400.40**
VarioPrep preparative columns		
10 mm ID	**736597.100**	**736400.40**
NUCLEOGEN® 500-7 DEAE		
Valco type analytical columns		
6 mm ID	**736598**	**736400.40**
VarioPrep preparative columns		
10 mm ID	**736599.100**	**736400.40**
NUCLEOGEN® 4000-7 DEAE		
Valco type analytical columns		
6 mm ID	**736601**	**736400.40**
VarioPrep preparative columns		
10 mm ID	**736602.100**	**736400.40**

NUCLEOGEN® ChromCart® guard column cartridges are 30 mm long, require the CC column holder 30 mm (REF 721823, see page 167) and are supplied in packs of 2. All other columns in packs of 1.

For information on DNA/RNA purification kits please ask for our catalogue "Bioanalysis"

HPLC columns for biochemical separations

NUCLEOGEL® SAX — anion exchange of biological macromolecules

- polymer-based strongly basic anion exchanger $-N^+(CH_3)_3$, gel matrix quaternised PEI; particle size 8 µm, pore size 1000 Å · USP **L23**
- pH working range 1 – 13, max. working pressure 200 bar
- eluent in column 0.1 M Na_2SO_4 + 0.2 % NaN_3
- recommended application:
  purification of peptides, large proteins and oligonucleotides, high capacity for proteins even at pH 10

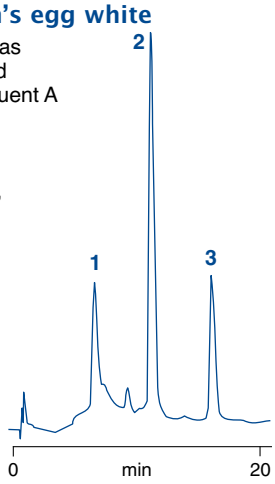
Ordering information

Pore size		Length →	50 mm	Guard columns
NUCLEOGEL® SAX				
Valco type analytical columns				
	1000 Å	4.6 mm ID	**719469**	**719600**
		7.7 mm ID	**719471**	**719600**

Separation of hen's egg white

Sample: frozen egg white was thawed, filtered and diluted 1 : 8 with eluent A
Column: 50 x 4.6 mm NUCLEOGEL® SAX 1000-8
Eluents: A) 0.01 M Tris-HCl, pH 7.5
B) A + 0.5 M NaAc, pH 7.5
Gradient: linear, 0 – 100 % B in 20 min
Flow rate: 1 ml/min
Inj. volume: 50 µl
Detection: UV, 280 nm

Peaks:
1. Conalbumin
2. Ovalbumin
3. not identified

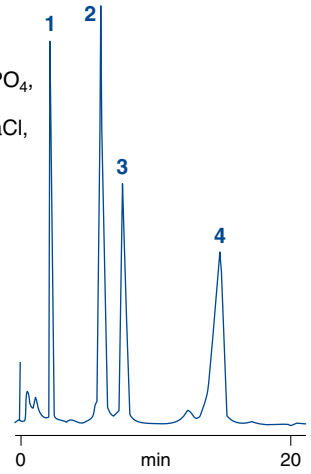


MN Appl. No. 115200

Separation of protein standards

Column: 50 x 4.6 mm NUCLEOGEL® SCX 1000-8
Eluents: A) 0.02 M KH_2PO_4, pH 6.0
B) A + 0.5 M NaCl, pH 6.0
Gradient: linear, 0 – 100 % B in 20 min
Flow rate: 1 ml/min
Detection: UV, 280 nm

Peaks:
1. Myoglobin
2. α-Chymotrypsinogen A
3. Cytochrome C
4. Lysozyme



MN Appl. No. 108260

NUCLEOGEL® SCX — cation exchange of biological macromolecules

- polymer-based strongly acidic cation exchanger $-SO_3^-$, hydrophilic gel matrix; particle size 8 µm, pore size 1000 Å · USP **L22**
- pH working range 1 – 13, max. working pressure 200 bar
- eluent in column 0.1 M Na_2SO_4 + 0.2 % NaN_3
- recommended application: proteins, peptides and carbohydrates with high isoelectric point

Ordering information

Pore size		Length →	50 mm	Guard columns
NUCLEOGEL® SCX				
Valco type analytical columns				
	1000 Å	4.6 mm ID	**719475**	**719540**
		7.7 mm ID	**719477**	**719540**

NUCLEOGEL® SAX and SCX Valco type guard columns measure 5 x 3 mm and require the guard column holder B, REF 719539, see page 169 (guard columns in packs of 2, columns in packs of 1).

NUCLEOSIL® MPN — RP chromatography of biological macromolecules

- silica-based reversed phase materials with monomerically bonded alkyl chains, brush type structure predominantly hydrophobic forces with a small portion of hydrophilic interactions
- **NUCLEOSIL® 100-5 C_{18} MPN:** octadecyl phase, particle size 5 µm, pore size 100 Å · USP **L1**
  dynamic protein binding capacity per g packing: 6 mg BSA, 110 mg cytochrome C
- **NUCLEOSIL® 120-3 C_{18} MPN:** octadecyl phase, particle size 3 µm, pore size 120 Å · USP **L1**
  dynamic protein binding capacity per g packing: 16 mg BSA, 55 mg cytochrome C
  outstanding selectivity for peptides
- **NUCLEOSIL® 300-5 C_4 MPN:** butyl phase, particle size 5 µm, pore size 300 Å · USP **L26**
  dynamic protein binding capacity per g packing: 14 mg BSA, 27 mg cytochrome C
  especially suited for the purification of larger, hydrophobic peptides and very different proteins
- pH working range 2 – 8, max. working pressure 250 bar
- maximum separation efficiency can be achieved when the injected protein mass does not exceed 1 – 2 % of the maximum protein loading capacity
- eluent in column methanol

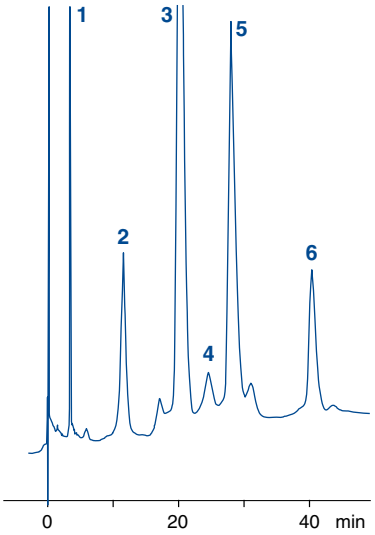
Separation of haemoglobin chains

Column:	250 x 4 mm NUCLEOSIL® 300-5 C_4 MPN
Eluents:	A) 20 % acetonitrile, 80 % water, 0.1 % TFA
	B) 60 % acetonitrile, 40 % water, 0.1 % TFA
Gradient:	40 – 60 % B in 60 min
Flow rate:	1 ml/min
Detection:	UV, 220 nm

Peaks:

1. Hem
2. β-globin
3. α-globin
4. $^{A}\gamma^{T}$-globin
5. $^{G}\gamma$-globin
6. $^{A}\gamma^{I}$-globin

MN Appl. No. 108240



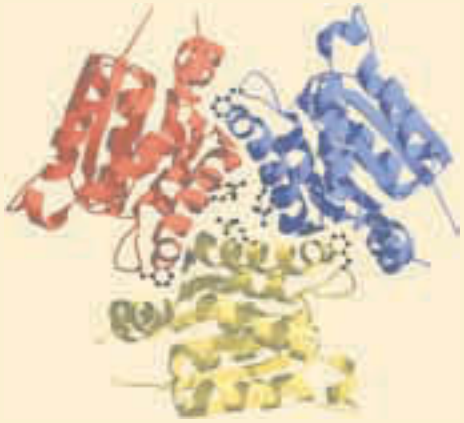
Ordering information

Length →	50 mm	125 mm	250 mm	Guard columns
EC analytical columns				
NUCLEOSIL® 100-5 C_{18} MPN				
4 mm ID		720230.40	720231.40	
NUCLEOSIL® 120-3 C_{18} MPN				
4 mm ID		720232.40		
NUCLEOSIL® 300-5 C_4 MPN				
4 mm ID	720244.40	720045.40	720245.40	721113.40

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
Guard columns in packs of 2, EC columns in packs of 1.

HPLC columns for biochemical separations

NUCLEOSIL® PPN



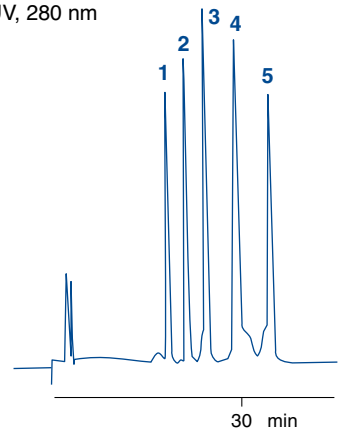
RP chromatography of biological macromolecules

- silica-based reversed phase materials with polymerically bonded alkyl chains; exclusively hydrophobic interactions
- **NUCLEOSIL® 100-5 C_{18} PPN:** octadecyl phase, particle size 5 µm, pore size 100 Å · USP **L1**
  dynamic protein binding capacity per g packing: 8 mg BSA, 64 mg cytochrome C; suited for the separation of peptides and proteins up to about 40 kD, also suited for basic peptides
- **NUCLEOSIL® 500-5 C_{18} PPN:** octadecyl phase, particle size 5 µm, pore size 500 Å · USP **L1**
  dynamic protein binding capacity per g packing: 22 mg BSA, 40 mg cytochrome C; especially suited for large peptides and medium-size hydrophilic proteins
- pH working range 1 – 9, max. working pressure 250 bar
- eluent in column methanol

Separation of a protein standard

Column: 125 x 4 mm NUCLEOSIL® 100-5 C_{18} PPN
Eluents: A) 0.1 % TFA in H_2O
B) 0.08 % TFA in CH_3CN
Gradient: 20 – 60 % B in 10 min
Flow rate: 1.0 ml/min
Detection: UV, 280 nm

Peaks:
1. Ribonuclease
2. Cytochrome C
3. Lysozyme

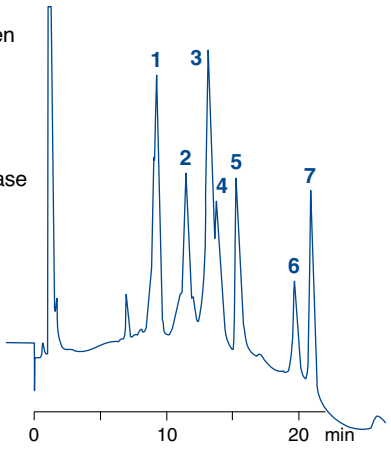


MN Appl. No. 108220

Separation of pancreatic secretion of piglets

Column: 125 x 4 mm NUCLEOSIL® 500-5 C_{18} PPN
Eluents: A) 0.1 % TFA in H_2O
B) 0.08 % TFA in CH_3CN
Gradient: linear 30 – 50 % B in 14 min, then 50 – 65 % B in 6 min
Flow rate: 1 ml/min
Detection: UV, 215 nm

Peaks:
1. Trypsin + trypsinogen
2. Proelastase
3. Lipase
   + α-chymotrypsin
4. Chymotrypsinogen
5. α-Amylase
6., 7. Procarboxypeptidase



MN Appl. No. 108280

Ordering information

Length →	50 mm	125 mm	250 mm	Guard columns
EC analytical columns				
NUCLEOSIL® 100-5 C_{18} PPN				
4 mm ID	**720250.40**	**720251.40**	**720252.40**	**721594.40**
NUCLEOSIL® 500-5 C_{18} PPN				
4 mm ID	**720256.40**	**720257.40**	**720258.40**	**721687.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359).
Guard columns in packs of 2, EC columns in packs of 1.

NUCLEOGEL® RP columns

RP columns for biochemical applications

- polystyrene resin cross-linked with divinylbenzene, available particle sizes 5 µm and 8 µm, available pore sizes 100 Å, 300 Å, 1000 Å and 4000 Å · USP L21
  pH working range 1 – 13, max. working pressure 180 bar
- small pore columns for reversed phase separation of small molecules such as pharmaceuticals with basic properties, e.g. organic heterocycles
  also suited for separation of nucleosides and nucleotides up to 5000 daltons
  allow gradient as well as isocratic elution
- wide pore columns are especially recommended for large biomolecules
  higher background hydrophobicity compared to silica phases
- eluent in column acetonitrile / water

Ordering information

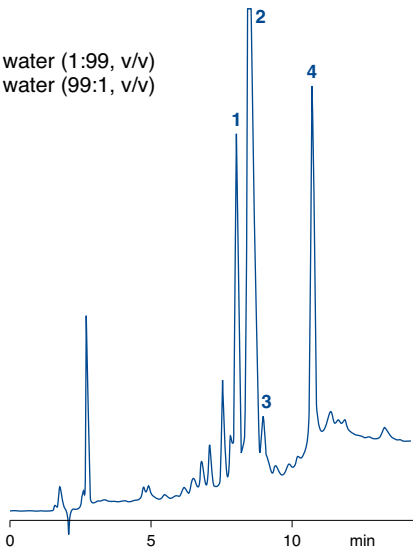
Length →	50 mm	150 mm	250 mm	300 mm	Guard columns
Valco type analytical columns 					
NUCLEOGEL® RP 100-5				pore size 100 Å, particle size 5 µm	
4.6 mm ID		**719454**	**719455**		**719542**
NUCLEOGEL® RP 100-8				pore size 100 Å, particle size 8 µm	
4.6 mm ID		**719456**	**719520**		**719542**
7.7 mm ID				**719457**	**719542**
NUCLEOGEL® RP 300-5				pore size 300 Å, particle size 5 µm	
4.6 mm ID	**719459**				**719542**
NUCLEOGEL® RP 300-8				pore size 300 Å, particle size 8 µm	
4.6 mm ID	**719460**				**719542**
7.7 mm ID	**719463**				**719542**

NUCLEOGEL® RP Valco type guard columns measure 5 x 3 mm and require the guard column holder B, REF 719539, see page 169. Guard columns in packs of 2, Valco type columns in packs of 1.

Analysis of the synthetic acyl carrier protein ACP(65-74)

Column: 150 x 4.6 mm
NUCLEOGEL® RP 100-8
Eluents: A) 0.1 % TFA in acetonitrile – water (1:99, v/v)
B) 0.1 % TFA in acetonitrile – water (99:1, v/v)
Gradient: 10 – 60 % B in 20 min
Flow rate: 1 ml/min
Detection: UV, 220 nm

Peaks:
1. ACP(66-74)(H-Gln)
2. ACP(65-74)
3. ACP(66-74)(Glp)
4. Thioanisole



MN Appl. No. 108500

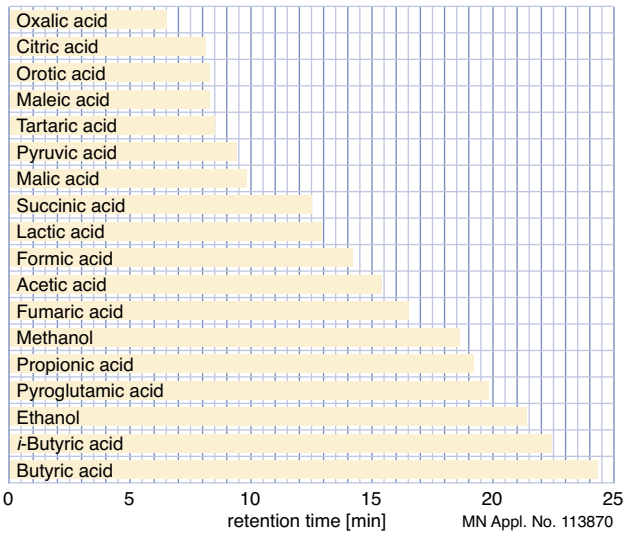
HPLC columns for sugar analysis

NUCLEOGEL® SUGAR 810 columns — separation of sugars

- sulphonated polystyrene / divinylbenzene resins in different ionic forms
  due to a different selectivity pattern compared to NUCLEOGEL® SUGAR columns, the range of application is considerably enlarged
- separation mechanism includes ion exclusion, ion exchange, size exclusion, ligand exchange as well as NP and RP chromatography
- H^+ form: separation of sugars, sugar alcohols and organic acids · USP **L17**
  eluent in column 0.01 N H_2SO_4
- Ca^{2+} form: separation of mono-, di- and oligosaccharides · USP **L19** · eluent in column water

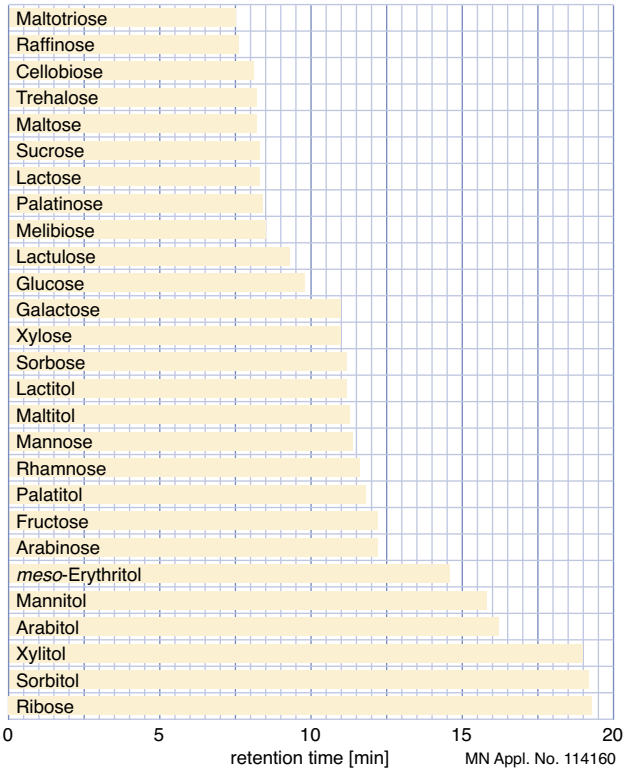
Organic acids and alcohols

Column: 300 x 7.8 mm NUCLEOGEL® SUGAR 810 H
Injection volume: 5 µl
Eluent: 5 mmol H_2SO_4
Flow rate: 0.6 ml/min
Temperature: 35 °C
Detection: RI



Sugars and sugar alcohols

Column: 300 x 7.8 mm NUCLEOGEL® SUGAR 810 Ca
Eluent: water , flow rate 0.6 ml/min
Detection: RI



Ordering information

	Length →	300 mm	Guard columns
Valco type columns			
NUCLEOGEL® SUGAR 810 H			
7.8 mm ID		**719574**	**719575**
NUCLEOGEL® SUGAR 810 Ca			
7.8 mm ID		**719570**	**719571**

NUCLEOGEL® SUGAR 810 ChromCart® guard columns measure 30 x 4 mm and require the ChromCart® column holder 30 mm, REF 721823, see page 167. They are supplied in packs of 2, all Valco type columns in packs of 1.

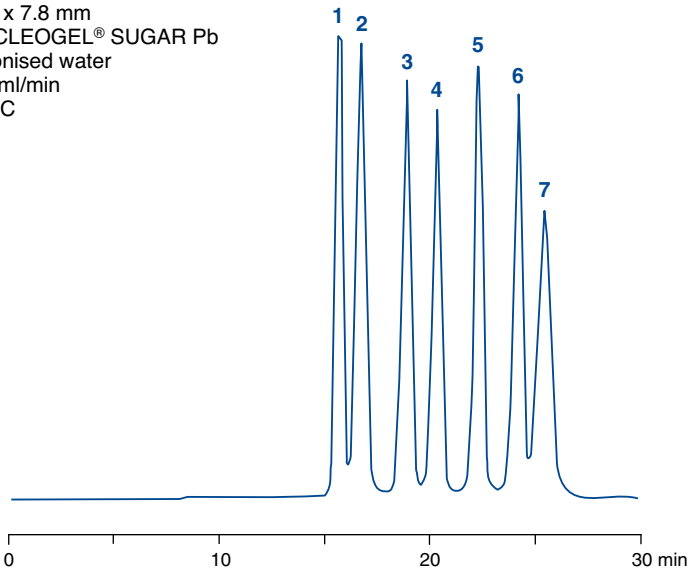
NUCLEOGEL® ION 300 OA / SUGAR columns — separation of sugars

- sulphonated spherical PS/DVB resins in different ionic forms; mean particle size 10 µm, pore size 100 Å
- separation mechanism includes steric exclusion, ligand exchange and partition effects, ligand exchange being the predominant force, since the hydrated metal ions form strong interactions with the hydroxyl groups of the sample molecules. The intensity of these interactions decreases in the sequence Pb, Ca, Na
- NUCLEOGEL® ION 300 OA: H^+ form for separation of sugars, alcohols and organic acids · USP **L17** eluent in column 0.01 N H_2SO_4
  - · Ca^{2+} form: separation of mono- and oligosaccharides, sugar alcohols · USP **L19**
  - · Na^+ form: separation of oligosaccharides from starch hydrolysates and food · USP **L58**
  - · Pb^{2+} form: separation of mono- and disaccharides from food and biological samples · USP **L34**
- eluent in column for Ca, Na and Pb phases: water + 0.02 % azide
- recommended operating temperatures: 60 – 95 °C; maximum pressure 100 bar

Separation of carbohydrates

Column: 300 x 7.8 mm NUCLEOGEL® SUGAR Pb
Eluent: deionised water
Flow rate: 0.4 ml/min
Temperature: 80 °C
Detection: RI

Peaks:
1. Sucrose
2. Maltose
3. Glucose
4. Xylose
5. Galactose
6. Arabinose
7. Mannose



MN Appl. No. 102430

Ordering information

Length →	300 mm	Guard columns
Valco type columns 		
NUCLEOGEL® ION 300 OA		
7.8 mm ID	**719501**	**719537**
NUCLEOGEL® SUGAR Ca		
6.5 mm ID	**719531**	**719535**
NUCLEOGEL® SUGAR Pb		
7.8 mm ID	**719530**	**719534**
NUCLEOGEL® SUGAR Na		
7.8 mm ID	**719532**	**719536**

NUCLEOGEL® ION and SUGAR Valco type guard columns measure 21 x 4 mm and require the guard column holder C (REF 719538, see page 169). Guard columns in packs of 2, Valco type columns in packs of 1.

HPLC columns for sugar analysis

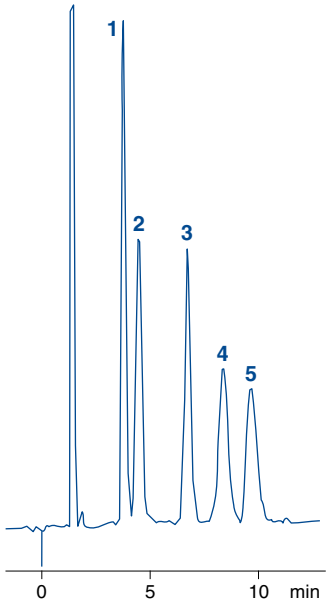
NUCLEOSIL® Carbohydrate — separation of mono- and disaccharides

- matrix: NUCLEOSIL® silica with amino modification, particle size 10 µm · USP **L8**
- recommended application: RP separation of mono- and disaccharides
- eluent in column acetonitrile / water (79:21, v/v)

Separation of sugars

Column:	250 x 4 mm NUCLEOSIL® Carbohydrate
Eluent:	acetonitrile – water (79:21, v/v)
Flow rate:	2 ml/min
Temperature:	25 °C
Detection:	RI
Injection volume:	10 µl

Peaks:
1. Fructose
2. Glucose
3. Saccharose
4. Maltose
5. Lactose



MN Appl. No. 102480

For the separation of oligosaccharides with longer chains ($10 < n < 40$) our phase NUCLEOSIL® 300-5 C_{18} can be successfully applied (see Application No. 102730 at *www.mn-net.com*). In this case a very flat gradient allows good resolution of the carbohydrates. For ordering information of this phase please see page 134.

Ordering information

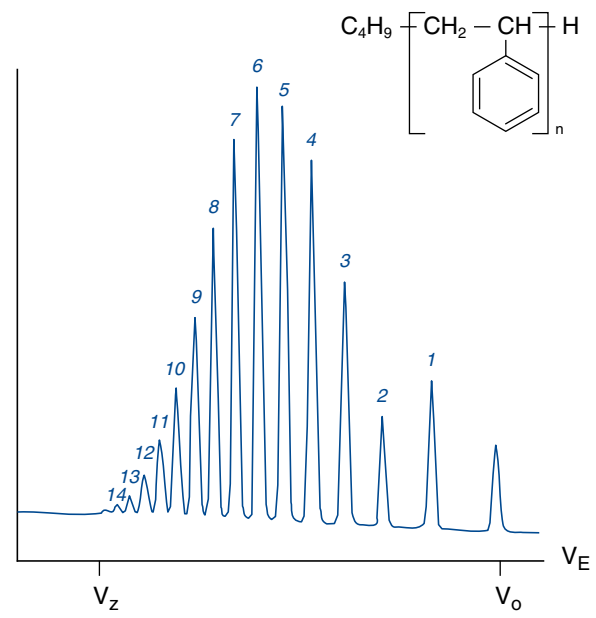
Length →	250 mm	Guard columns
NUCLEOSIL® Carbohydrate		
EC columns		
4 mm ID	**720905.40**	**721595.40**

As guard columns for EC columns use ChromCart® guard column cartridges with guard column adaptor EC (REF 721359). Columns and guard columns in packs of 1.

NUCLEOGEL® GPC — for GPC of water-insoluble substances

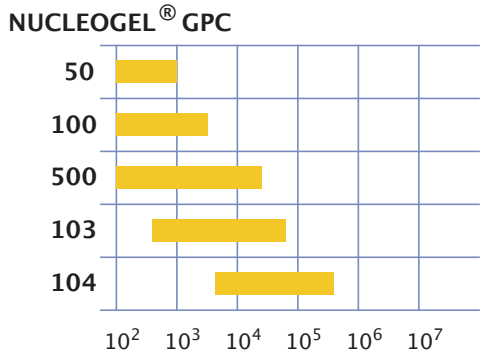
- highly crosslinked macroporous, spherical polystyrene – divinylbenzene polymer matrix with good mechanical stability
- eluent in column toluene

Chromatogram of styrene oligomers



exclusion volume — dead time

Working ranges for polystyrene



Ordering information

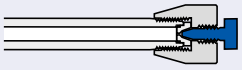
Phase	Exclusion limit [kDaltons]	Application	Column 300 x 7.7 mm
Valco type analytical columns			
5 µm particles			
NUCLEOGEL GPC 50-5	2	low molecular weight organics	**719402**
NUCLEOGEL GPC 100-5	4	oligomers, oils	**719403**
NUCLEOGEL GPC 500-5	25	low molecular weight polymers	**719404**
NUCLEOGEL GPC 103-5	60	low molecular weight polymers	**719405**
NUCLEOGEL GPC 104-5	500	polymers up to 500 kDaltons	**719406**
		guard column 50 x 7.7 mm	**719409**
10 µm particles			
NUCLEOGEL GPC 50-10	2	low molecular weight organics	**719410**
NUCLEOGEL GPC 100-10	4	oligomers, oils	**719411**
NUCLEOGEL GPC 500-10	25	low molecular weight polymers	**719412**
NUCLEOGEL GPC 103-10	60	low molecular weight polymers	**719413**
NUCLEOGEL GPC 104-10	500	polymers up to 500 kDaltons	**719414**
		guard column 50 x 7.7 mm	**719418**

Columns and guard columns in packs of 1.

EC standard columns for analytical HPLC

- analytical column system manufactured from stainless steel
  M 8 outer threads on both ends
  combination of sealing element and very fine-meshed stainless steel screen, PTFE ring and fitting adaptor
  column heads SW 12, with inner threads M 8 and UNF 10-32
- as built-in guard columns ChromCart® guard column cartridges with 8 mm length are used with the guard column adaptor EC (see below)
- supplied with NUCLEODUR® and NUCLEOSIL® spherical silicas

Available standard dimensions of EC columns · please ask for availability of certain phases

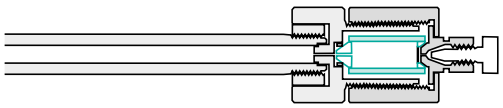
ID [mm]	Length [mm] 8*	20	30	50	75	100	125	150	200	250	300	End fitting design
2	-	x	x	x	x	x	x	x	x	x	x	
3	x	x	x	x	x	x	x	x	x	x	x	
4	x	x	x	x	x	x	x	x	x	x	x	
4.6	-	x	x	x	x	x	x	x	x	x	x	

* Please note that 3 mm ID ChromCart® guard column cartridges are applicable for 2 mm and 3 mm ID EC columns, and 4 mm ID guard column cartridges are used for 4 mm and 4.6 mm ID EC columns.

Installation of the EC guard column adaptor




EC column with CC guard column



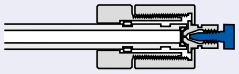
Accessories and replacement parts for EC columns · Ordering information

Description	Pack of	REF
Guard column adaptor EC	1	**721359**
1/16“ nut for connecting 1/16“ capillaries	5	**718583**
1/16“ ferrule	5	**718584**
1/16“ end cap, plastic	4	**718582**
EC fitting adaptor	1	**718987**
EC column head (nut)	1	**718988**
EC PTFE sealing ring	4	**718992**
3-part sealing combination for EC columns	5 kits	**718998**

ChromCart® cartridge system

- analytical column system manufactured from stainless steel (US patent 5,342,515)
- rapid and convenient installation
  columns are changed without removal of capillary connections
  all unions are screwed by hand
  easy installation of guard cartridges without special adaptor
  connection of columns of different lengths and inner diameters with one type of connecting kit (see below)
- supplied with NUCLEOSIL® spherical silicas as well as with well-known packings from other manufacturers

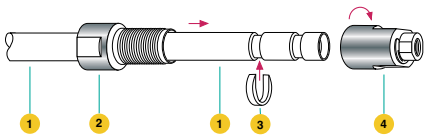
Available standard dimensions of ChromCart® cartridges · please ask for availability of certain phases

ID [mm]	Length [mm]				End fitting design
	8*	125	150	250	
2	-	x	x	x	
3	x	x	x	x	
4	x	x	x	x	
4.6	-	x	x	x	

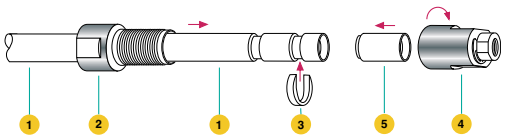
* Please note that 3 mm ID guard column cartridges are also applicable for 2 mm ID CC columns, and 4 mm ID guard column cartridges are also used for 4.6 mm ID CC columns.

Connection of ChromCart® cartridges and guard column cartridges

Use of 1 analytical column



Use of column and guard column



Legend

1. analytical column
2. sleeve
3. guide ring
4. nut
5. guard column
6. PEEK sealing cone
7. longer nut

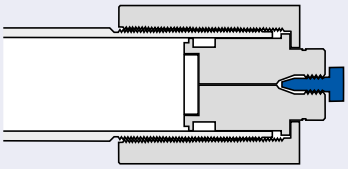
Accessories for the ChromCart® cartridge system · Ordering information

Description	Pack of	REF
CC connecting kit (consists of 2 nuts with end fittings, two sleeves and two guide rings)	1 kit	**721690**
CC nut with end fitting	1 set	**721691**
CC sleeve with outer threads	1	**721692**
CC guide ring	1	**721693**
CC coupling kit (consists of longer nut, PEEK seal, sleeve with outer threads and 2 guide rings for coupling two CC columns)	1 kit	**721694**
PEEK seal	2	**721695**
CC guard column holder 8 mm for stand-alone operation of 8 mm CC cartridges	1	**721820**
CC column holder 30 mm for stand-alone operation of 30 mm CC cartridges	1	**721823**

VarioPrep columns (VP)

- column system for preparative HPLC manufactured from stainless steel with adjustable end fittings (suitable for frequent use of back-flushing techniques)
- allows compensation of a dead volume, which could result at the column inlet after some time of operation, without need for opening the column
- supplied with NUCLEODUR® and NUCLEOSIL® spherical silicas
- up-scaling from analytical to preparative columns see page 172

Available standard dimensions of VarioPrep columns with axially adjustable end fitting

ID [mm]	Length [mm]									End fitting design
	10*	15*	20*	50	100	125	150	250	500	
8	x			x	x	x	x	x		
10				x	x	x	x	x		
16			x	x	x	x	x	x		
21				x	x	x	x	x		
32		x			x		x	x		
40				x	x	x	x	x	x	
50					x		x	x		
80								x	x	

10 x 8 mm guard columns for 8 and 10 mm ID VP columns, 20 x 16 mm guard columns for 16 and 21 mm ID VP columns, 15 x 32 mm guard columns for 32 to 50 mm ID VP columns. VP guard columns require the holders listed below.

VarioPrep guard column holders and replacement parts · ordering information

Description	Pack of	REF
VP guard column holder 8 mm for VarioPrep columns with 8 and 10 mm ID	1	**718251**
O-ring for VP guard column holder 8 mm	2	**718975**
VP guard column holder 16 mm for VarioPrep columns with 16 and 21 mm ID	1	**718250**
O-ring for VP guard column holder 16 mm	2	**718976**
VP guard column holder 32 mm for VarioPrep columns with 32 to 50 mm ID	1	**718253**
O-ring for VP guard column holder 32 mm	2	**718977**

Replacement parts for VarioPrep columns · Ordering information

Description	Pack of	REF
for VarioPrep columns with 10 mm ID		
VP plunger fitting 10 mm	1	**718837**
VP nut 10 mm	1	**718842**
VP sealing element set 10 mm	1 set	**718931**
VP sealing ring set 10 mm	1 set	**718852**
VP MN Inert sealing combination 10 mm	1 set	**718848**
for VarioPrep columns with 21 mm ID		
VP plunger fitting 21 mm	1	**718861**
VP nut 21 mm	1	**718862**
VP sealing element set 21 mm	1 set	**718853**
VP sealing ring set 21 mm	1 set	**718854**
VP MN Inert sealing combination 21 mm	1 set	**718870**

Microbore columns

- analytical column system for rapid HPLC and LC/MS analyses with high resolution
- available lengths: 40, 60, 100, 125, 150, 200, 250 and 300 mm, available IDs: 0.05, 0.075, 0.1, 0.15, 0.3, 0.4, 0.5, 0.75, 1, 1.5 mm
- Microbore columns up to 0.3 mm ID are fused silica capillaries, while microbore columns with 0.3 – 1.5 mm ID are stainless steel columns.
- supplied with NUCLEODUR® RP phases (NUCLEOSIL® on request)
- guard columns for microbore columns are available on request.

Advantages of microbore columns

- only small sample volumes required
- highest detection sensitivity
- low flow rate = reduced eluent consumption
- **time saving + reduced eluent consumption = reduced cost**

Change of flow rate and solvent saving as a function of the column inner diameter

ID [mm]	Flow rate [ml/min]	Solvent saving	Increase in sensitivity
4.6	1.3	-	-
4.0	1.0	~ 25 %	~ 1.3
3.0	0.56	~ 57 %	~ 2.4
2.0	0.25	~ 81 %	~ 5.3
1.0	0.06	~ 95 %	~ 21.7

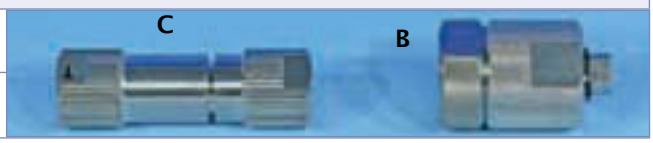
for a constant length relative to a column with 4.6 mm ID and a flow rate of 1.3 ml/min for isocratic application

Valco type columns

- analytical column system manufactured from stainless steel
  available inner diameters: 4.6 mm ID (1/4" OD) and 7.7 mm (3/8" OD)
- mainly used for some phases for special separations

Accessories for Valco type columns · Ordering information

Description	Pack of	REF
Guard column holder B for VA guard columns 5 x 3 mm	1	**719539**
Guard column holder C for VA guard columns 21 x 4 mm	1	**719538**



Frits 2 µm for 4.6 mm ID columns	5	**719485**
Frits 2 µm for 7.7 mm ID columns	5	**719486**
Column connecting nuts for 1/16" capillaries	5	**719487**
Ferrules for 1/16" capillaries	5	**719488**
Union for columns	1	**719489**
Column end plugs	5	**719490**

Accessories for stainless steel HPLC columns

- Stainless steel columns are most frequently used in HPLC. The material is corrosion resistant, pressure stable and easy to work mechanically.

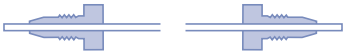
Ordering information

Stainless steel capillary tubing

Length		OD		ID	Pack of	REF
Capillary tubing in coils						
3 m	x	1/16"	x	0.25 mm	1 coil	**718737**
3 m	x	1/16"	x	0.5 mm	1 coil	**718738**
1 m	x	1/16"	x	0.12 mm	1 coil	**718790**
1 m	x	1/16"	x	0.25 mm	1 coil	**718735**
1 m	x	1/16"	x	0.5 mm	1 coil	**718736**

Capillary tubing, cut pieces, ready-to-use						
50 mm	x	1/16"	x	0.12 mm	2 tubes	**718731**
100 mm	x	1/16"	x	0.12 mm	2 tubes	**718732**
200 mm	x	1/16"	x	0.12 mm	2 tubes	**718733**
300 mm	x	1/16"	x	0.12 mm	2 tubes	**718734**
100 mm	x	1/16"	x	0.25 mm	5 tubes	**718588**
200 mm	x	1/16"	x	0.25 mm	5 tubes	**718635**
300 mm	x	1/16"	x	0.25 mm	5 tubes	**718589**
100 mm	x	1/16"	x	0.5 mm	5 tubes	**718516**
300 mm	x	1/16"	x	0.5 mm	5 tubes	**718517**
50 mm	x	1/32"	x	0.12 mm	2 tubes	**718670**
100 mm	x	1/32"	x	0.12 mm	2 tubes	**718671**
200 mm	x	1/32"	x	0.12 mm	2 tubes	**718672**
50 mm	x	1/32"	x	0.25 mm	2 tubes	**718673**
100 mm	x	1/32"	x	0.25 mm	2 tubes	**718674**
50 mm	x	1/32"	x	0.5 mm	2 tubes	**718676**
100 mm	x	1/32"	x	0.5 mm	2 tubes	**718677**
200 mm	x	1/32"	x	0.5 mm	2 tubes	**718678**

Stainless steel accessories

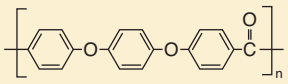
Description		Pack of	REF
Capillary accessories			
Knife file		1	**706121**
Cutter for 1/16" capillaries		1	**706290**
Capillary union 100 mm x 1/16" x 0.25 mm 		1	**718637**
Eluent filters, stainless steel			
for 1/16" tubing	2 µm frit	1	**718750**
for 1/16" tubing	10 µm frit	1	**718752**
for 1/8" tubing	2 µm frit	1	**718751**
for 1/8" tubing	10 µm frit	1	**718753**

For accessories and replacement parts for EC columns see page 166, for accessories and replacement parts for ChromCart® cartridges see page 167, replacement parts for VarioPrep columns are listed on page 168.

PEEK accessories

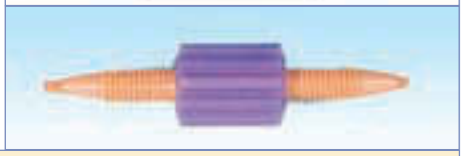
- PEEK (= polyether ether ketone) is a high performance polymer belonging to the group of polyarylether ketones (PAEK), which meets all requirements of HPLC columns with respect to chemical resistance and mechanical stability. In some fields of application in HPLC, like e.g. in ion chromatography and chromatography of biopolymers, PEEK fulfils the requirements for a nonmetallic material.

PEEK



- All fittings can be tightened by hand. The following table summarizes the available PEEK products.

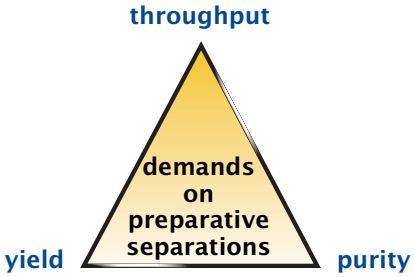
Ordering information

Description	Pack of	REF	
PEEK fittings			
1/16" PEEK fingertight fitting, 1-part combination nut + ferrule	1	**718770**	
1/16" PEEK fingertight nut	1	**718771**	
1/16" PEEK ferrule for REF 718771	1	**718772**	
1/16" PEEK double ferrule	1	**718775**	
1/16" PEEK union, both sides inner threads, equipped with 2 fingertight nuts and double ferrules	1	**718766**	
1/16" PEEK union, both sides inner threads, however without nuts and without ferrules	1	**718767**	
1/16" PEEK union, both sides outer threads	1	**718768**	
PEEK standard capillaries			
OD / ID [mm] / Length			
1/16" / 0.13 / 1 m	1	**718765**	
1/16" / 0.17 / 1 m	1	**718760**	
1/16" / 0.25 / 1 m	1	**718761**	
1/16" / 0.5 / 1 m	1	**718762**	
1/16" / 0.75 / 1 m	1	**718763**	
Tools for PEEK capillaries			
Guillotine cutter for PEEK and PTFE capillaries	1	**718769**	
Clean-Cut cutter for different capillary outer diameters	1	**718755**	

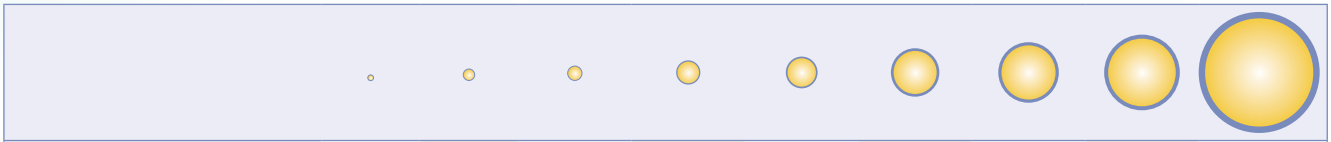
NUCLEODUR® high purity silica for HPLC

Basic rules of preparative HPLC

Basically, preparative HPLC follows the same rules as analytical scale chromatography. However, there are important differences in the aims of the two techniques. In analytical HPLC chromatographers focus on peak shape, and resolution of all eluted analytes, whereas in preparative chromatography yield and purity of the final product, as well as cost-effectiveness of the method, are emphasised.



Scale up factors and parameters for typical MN column dimensions



ID x length [mm]	4 x 250	8 x 250	10 x 250	16 x 250	21 x 250	32 x 250	40 x 250	50 x 250	80 x 250
Linear scale-up factor	1	4	6.25	16	27.6	64	100	161.3	400
Typical sample mass* [mg]	0.02 – 2	0.08 – 8	0.13 – 13	0.3 – 35	0.6 – 60	1.3 – 130	2 – 210	3 – 350	10 – 850
Typical flow rate [ml/min]	0.5 – 1.5	2 – 6	3 – 9	8 – 24	14 – 40	32 – 96	50 – 150	80 – 250	200 – 600

* For RP material; the maximum amounts given here always depend on the separation problem and on the sample composition. In some cases half of the amount given can cause drastic overload, in other cases the maximum amounts can be even higher still giving acceptable separations.

NUCLEODUR® bulk packings

- totally spherical high purity silica
- pore size 110 Å, pore volume 0.9 ml/g, surface (BET) 340 m^2/g, density 0.47 g/ml, pressure stability 800 bar
- larger particles for preparative applications

Ordering information

Phase	Endcapped	Carbon content	Particle size	Pack of 100 g	Pack of 1000 g
NUCLEODUR® C_{18} HTec premium octadecyl phases (see p. 118)					
NUCLEODUR® 100-5 C_{18} HTec	yes	18 % C	5 µm	**713830.0100**	**713830.1**
NUCLEODUR® 100-7 C_{18} HTec	yes	18 % C	7 µm	**713831.0100**	**713831.1**
NUCLEODUR® 100-10 C_{18} HTec	yes	18 % C	10 µm	**713832.0100**	**713832.1**
NUCLEODUR® C_{18} ec standard octadecyl phases (see p. 120)					
NUCLEODUR® 100-10 C_{18} ec	yes	17.5 % C	10 µm	**713611.0100**	**713611.1**
NUCLEODUR® 100-12 C_{18} ec	yes	17.5 % C	12 µm	**713618.0100**	**713618.1**
NUCLEODUR® 100-16 C_{18} ec	yes	17.5 % C	16 µm	**713621.0100**	**713621.1**
NUCLEODUR® 100-20 C_{18} ec	yes	17.5 % C	20 µm	**713601.0100**	**713601.1**
NUCLEODUR® 100-30 C_{18} ec	yes	17.5 % C	30 µm	**713631.0100**	**713631.1**
NUCLEODUR® 100-50 C_{18} ec	yes	17.5 % C	50 µm	**713550.0100**	**713550.1**
Unmodified NUCLEODUR® silica (see p. 123)					
NUCLEODUR® 100-10			10 µm	**713610.0100**	**713610.1**
NUCLEODUR® 100-12			12 µm	**713615.0100**	**713615.1**
NUCLEODUR® 100-16			16 µm	**713620.0100**	**713620.1**
NUCLEODUR® 100-20			20 µm	**713600.0100**	**713600.1**
NUCLEODUR® 100-30			30 µm	**713630.0100**	**713630.1**
NUCLEODUR® 100-50			50 µm	**713551.0100**	**713551.1**

NUCLEOSIL® bulk packings

- spherical silica
- pH stability 2 – 8 (for NUCLEOSIL® 100-5 C_{18} AB 1 – 9)
- for a characterisation of our NUCLEOSIL® silica see page 101

Physical properties of unmodified NUCLEOSIL® materials

Phase	Pore size	Pore volume	Surface (BET)	Density	Pressure stability
NUCLEOSIL® 50	50 Å	0.8 ml/g	420 m^2/g	0.45 g/ml	600 bar
NUCLEOSIL® 100	100 Å	1 ml/g	350 m^2/g	0.36 g/ml	600 bar
NUCLEOSIL® 120	120 Å	0.65 ml/g	200 m^2/g	0.55 g/ml	800 bar
NUCLEOSIL® 300	300 Å	0.8 ml/g	100 m^2/g	0.45 g/ml	400 bar
NUCLEOSIL® 500	500 Å	0.8 ml/g	35 m^2/g	0.45 g/ml	400 bar
NUCLEOSIL® 1000	1000 Å	0.8 ml/g	25 m^2/g	0.45 g/ml	300 bar
NUCLEOSIL® 4000	4000 Å	0.7 ml/g	10 m^2/g	0.48 g/ml	300 bar

For description of individual modifications see chapter “Columns with NUCLEOSIL®” from page 130.

Ordering information

Phase	Endcapped	Carbon content	Pore size	Particle size	Pack of 10 g	Pack of 100 g
Octadecyl phases						$-(CH_2)_{17}-CH_3$
NUCLEOSIL® 50-5 C_{18} ec	yes	14.5 % C	50 Å	5 µm	**712031.10**	**712031.100**
NUCLEOSIL® 100-5 C_{18} AB	yes	24 % C	100 Å	5 µm	**712952.10**	**712952.100**
NUCLEOSIL® 100-3 C_{18}	yes	15 % C	100 Å	3 µm	**712370.10**	**712370.100**
NUCLEOSIL® 100-5 C_{18}	yes	15 % C	100 Å	5 µm	**712130.10**	**712130.100**
NUCLEOSIL® 100-7 C_{18}	yes	15 % C	100 Å	7 µm	**712140.10**	**712140.100**
NUCLEOSIL® 100-10 C_{18}	yes	15 % C	100 Å	10 µm	**712150.10**	**712150.100**
NUCLEOSIL® 120-3 C_{18}	yes	11 % C	120 Å	3 µm	**712460.10**	**712460.100**
NUCLEOSIL® 120-5 C_{18}	yes	11 % C	120 Å	5 µm	**712470.10**	**712470.100**
NUCLEOSIL® 120-7 C_{18}	yes	11 % C	120 Å	7 µm	**712480.10**	**712480.100**
NUCLEOSIL® 120-10 C_{18}	yes	11 % C	120 Å	10 µm	**712490.10**	**712490.100**
NUCLEOSIL® 300-5 C_{18}	yes	6.5 % C	300 Å	5 µm	**712520.10**	**712520.100**
NUCLEOSIL® 300-7 C_{18}	yes	6.5 % C	300 Å	7 µm	**712530.10**	**712530.100**
NUCLEOSIL® 300-10 C_{18}	yes	6.5 % C	300 Å	10 µm	**712540.10**	**712540.100**
NUCLEOSIL® 500-7 C_{18}	yes	2 % C	500 Å	7 µm	**712760.10**	**712760.100**
NUCLEOSIL® 1000-7 C_{18}	yes	~ 1 % C	1000 Å	7 µm	**712790.10**	**712790.100**
NUCLEOSIL® 4000-7 C_{18}	yes	<1 % C	4000 Å	7 µm	**712926.10**	**712926.100**
Octyl phases						$-(CH_2)_7-CH_3$
NUCLEOSIL® 50-5 C_8 ec	yes	9 % C	50 Å	5 µm	**712032.10**	**712032.100**
NUCLEOSIL® 100-5 C_8 ec	yes	9 % C	100 Å	5 µm	**712101.10**	**712101.100**
NUCLEOSIL® 100-5 C_8	no	8.5 % C	100 Å	5 µm	**712100.10**	**712100.100**
NUCLEOSIL® 100-7 C_8	no	8.5 % C	100 Å	7 µm	**712110.10**	**712110.100**
NUCLEOSIL® 100-10 C_8	no	8.5 % C	100 Å	10 µm	**712120.10**	**712120.100**
NUCLEOSIL® 120-3 C_8	no	6.5 % C	120 Å	3 µm	**712570.10**	**712570.100**
NUCLEOSIL® 120-5 C_8	no	6.5 % C	120 Å	5 µm	**712580.10**	**712580.100**
NUCLEOSIL® 120-7 C_8	no	6.5 % C	120 Å	7 µm	**712500.10**	**712500.100**
NUCLEOSIL® 120-10 C_8	no	6.5 % C	120 Å	10 µm	**712590.10**	**712590.100**
NUCLEOSIL® 300-5 C_8	no	~ 3 % C	300 Å	5 µm	**712650.10**	**712650.100**
NUCLEOSIL® 300-7 C_8	no	~ 3 % C	300 Å	7 µm	**712550.10**	**712550.100**
NUCLEOSIL® 300-10 C_8	no	~ 3 % C	300 Å	10 µm	**712660.10**	**712660.100**
NUCLEOSIL® 500-7 C_8	no	<1 % C	500 Å	7 µm	**712830.10**	**712830.100**

NUCLEOSIL® standard silica for HPLC

Phase	Endcapped	Carbon content	Pore size	Particle size	Pack of 10 g	Pack of 100 g
Phenyl phases						**$-(CH_2)_3 - C_6H_5$**
NUCLEOSIL® 100-5 C_6H_5 ec	yes	8% C	100 Å	5 µm	712311.10	712311.100
NUCLEOSIL® 100-5 C_6H_5	no	8% C	100 Å	5 µm	712310.10	712310.100
NUCLEOSIL® 100-7 C_6H_5	no	8% C	100 Å	7 µm	712340.10	712340.100
NUCLEOSIL® 120-7 C_6H_5	no	6.5% C	120 Å	7 µm	712510.10	712510.100
NUCLEOSIL® 300-7 C_6H_5	no	~ 3% C	300 Å	7 µm	712670.10	712670.100
NUCLEOSIL® 500-7 C_6H_5	no	~ 2% C	500 Å	7 µm	712923.10	712923.100
NUCLEOSIL® 1000-7 C_6H_5	no	~ 1% C	1000 Å	7 µm	712924.10	712924.100
Butyl phases						**$-(CH_2)_3 - CH_3$**
NUCLEOSIL® 120-5 C_4	yes	~ 4% C	120 Å	5 µm	712290.10	712290.100
NUCLEOSIL® 300-5 C_4	yes	~ 2% C	300 Å	5 µm	712620.10	712620.100
NUCLEOSIL® 300-7 C_4	yes	~ 2% C	300 Å	7 µm	712630.10	712630.100
NUCLEOSIL® 300-10 C_4	yes	~ 2% C	300 Å	10 µm	712640.10	712640.100
NUCLEOSIL® 500-7 C_4	yes	<1% C	500 Å	7 µm	712750.10	712750.100
NUCLEOSIL® 1000-7 C_4	yes	<1% C	1000 Å	7 µm	712780.10	712780.100
NUCLEOSIL® 4000-7 C_4	yes	<1% C	4000 Å	7 µm	712925.10	712925.100
Dimethyl phases						**$-(CH_3)_2$**
NUCLEOSIL® 100-7 C_2	no	3.5% C	100 Å	7 µm	712080.10	712080.100
Cyano phases (nitrile)						**$-(CH_2)_3 - CN$**
NUCLEOSIL® 100-5 CN		5% C	100 Å	5 µm	712160.10	712160.100
NUCLEOSIL® 100-10 CN		5% C	100 Å	10 µm	712170.10	712170.100
NUCLEOSIL® 120-7 CN		~ 3% C	120 Å	7 µm	712600.10	712600.100
NUCLEOSIL® 300-7 CN		~ 2.5% C	300 Å	7 µm	712820.10	712820.100
NUCLEOSIL® 500-7 CN		~ 2% C	500 Å	7 µm	712840.10	712840.100
Nitro phases						**$-(CH_2)_3 - C_6H_4\ NO_2$**
NUCLEOSIL® 100-5 NO_2		~ 4.5% C	100 Å	5 µm	712180.10	712180.100
NUCLEOSIL® 100-10 NO_2		~ 4.5% C	100 Å	10 µm	712190.10	712190.100
Diol phases						**$-(CH_2)_3 - O - CH_2 - CH(OH) - CH_2OH$**
NUCLEOSIL® 100-7 OH (Diol)		5% C	100 Å	7 µm	712350.10	712350.100
NUCLEOSIL® 300-7 OH (Diol)		~ 1.5% C	300 Å	7 µm	712560.10	712560.100
NUCLEOSIL® 500-7 OH (Diol)		~ 1.5% C	500 Å	7 µm	712740.10	712740.100
NUCLEOSIL® 1000-7 OH (Diol)		~ 1% C	1000 Å	7 µm	712770.10	712770.100
NUCLEOSIL® 4000-7 OH (Diol)		~ 1% C	4000 Å	7 µm	712927.10	712927.100
Amino phases						**$-(CH_2)_3 - NH_2$**
NUCLEOSIL® 100-5 NH_2		3.5% C	100 Å	5 µm	712200.10	712200.100
NUCLEOSIL® 100-10 NH_2		3.5% C	100 Å	10 µm	712210.10	712210.100
NUCLEOSIL® 120-7 NH_2		~ 2% C	120 Å	7 µm	712610.10	712610.100
NUCLEOSIL® 300-7 NH_2		~ 2% C	300 Å	7 µm	712919.10	712919.100
Dimethylamino phases						**$-(CH_2)_3 - N(CH_3)_2$**
NUCLEOSIL® 100-5 $N(CH_3)_2$		4% C	100 Å	5 µm	712220.10	712220.100
NUCLEOSIL® 100-10 $N(CH_3)_2$		4% C	100 Å	10 µm	712230.10	712230.100
Cation exchanger, strongly acidic (SCX)						**$-(CH_2)_3 - C_6H_4 - SO_3\ Na$**
NUCLEOSIL® 100-5 SA		6.5% C	100 Å	5 µm	712240.10	712240.100
NUCLEOSIL® 100-10 SA		6.5% C	100 Å	10 µm	712250.10	712250.100

Phase	Endcapped	Carbon content	Pore size	Particle size	Pack of 10 g	Pack of 100 g
Anion exchanger, strongly basic (SAX)					$-(CH_2)_3-C_6H_4-CH_2-N^+(CH_3)_3Cl^-$	
NUCLEOSIL® 100-5 SB		10 % C	100 Å	5 µm	**712260.10**	**712260.100**
NUCLEOSIL® 100-10 SB		10 % C	100 Å	10 µm	**712270.10**	**712270.100**
Unmodified silica						**SiOH**
NUCLEOSIL® 50-3			50 Å	3 µm	**712000.10**	**712000.100**
NUCLEOSIL® 50-5			50 Å	5 µm	**712010.10**	**712010.100**
NUCLEOSIL® 50-7			50 Å	7 µm	**712020.10**	**712020.100**
NUCLEOSIL® 50-10			50 Å	10 µm	**712030.10**	**712030.100**
NUCLEOSIL® 100-3			100 Å	3 µm	**712360.10**	**712360.100**
NUCLEOSIL® 100-5			100 Å	5 µm	**712040.10**	**712040.100**
NUCLEOSIL® 100-7			100 Å	7 µm	**712050.10**	**712050.100**
NUCLEOSIL® 100-10			100 Å	10 µm	**712060.10**	**712060.100**
NUCLEOSIL® 120-3			120 Å	3 µm	**712390.10**	**712390.100**
NUCLEOSIL® 120-5			120 Å	5 µm	**712400.10**	**712400.100**
NUCLEOSIL® 120-7			120 Å	7 µm	**712410.10**	**712410.100**
NUCLEOSIL® 120-10			120 Å	10 µm	**712420.10**	**712420.100**
NUCLEOSIL® 300-5			300 Å	5 µm	**712430.10**	**712430.100**
NUCLEOSIL® 300-7			300 Å	7 µm	**712440.10**	**712440.100**
NUCLEOSIL® 300-10			300 Å	10 µm	**712450.10**	**712450.100**
NUCLEOSIL® 500-5			500 Å	5 µm	**712680.10**	**712680.100**
NUCLEOSIL® 500-7			500 Å	7 µm	**712690.10**	**712690.100**
NUCLEOSIL® 500-10			500 Å	10 µm	**712700.10**	**712700.100**
NUCLEOSIL® 1000-5			1000 Å	5 µm	**712710.10**	**712710.100**
NUCLEOSIL® 1000-7			1000 Å	7 µm	**712720.10**	**712720.100**
NUCLEOSIL® 1000-10			1000 Å	10 µm	**712730.10**	**712730.100**
NUCLEOSIL® 4000-5			4000 Å	5 µm	**712850.10**	**712850.100**
NUCLEOSIL® 4000-7			4000 Å	7 µm	**712860.10**	**712860.100**
NUCLEOSIL® 4000-10			4000 Å	10 µm	**712870.10**	**712870.100**

POLYGOSIL® bulk packings

- irregular silica for analytical applications
- pH stability 2 – 8

Physical properties of unmodified POLYGOSIL® materials

Phase	Pore size	Pore volume	Surface (BET)	Density	Pressure stability
POLYGOSIL® 60	60 Å	0.75 ml/g	350 m^2/g	0.45 g/ml	600 bar
POLYGOSIL® 100	100 Å	1 ml/g	280 m^2/g	0.35 g/ml	400 bar
POLYGOSIL® 300	300 Å	0.8 ml/g	100 m^2/g	0.45 g/ml	400 bar
POLYGOSIL® 1000	1000 Å	0.8 ml/g	25 m^2/g	0.45 g/ml	300 bar

Modification of POLYGOSIL® follows the same processes as for NUCLEOSIL® silica.

POLYGOSIL® irregular silica for HPLC

Ordering information

Phase	Endcapped	Carbon content	Pore size	Particle size	Pack of 10 g	Pack of 100 g
Octadecyl phases						**$-(CH_2)_{17}-CH_3$**
POLYGOSIL® 60-5 C_{18}	yes	12 % C	60 Å	5 µm	**711330.10**	**711330.100**
POLYGOSIL® 60-7 C_{18}	yes	12 % C	60 Å	7 µm	**711340.10**	**711340.100**
POLYGOSIL® 60-10 C_{18}	yes	12 % C	60 Å	10 µm	**711350.10**	**711350.100**
POLYGOSIL® 100-5 C_{18}	yes	14 % C	100 Å	5 µm	**711560.10**	**711560.100**
POLYGOSIL® 100-7 C_{18}	yes	14 % C	100 Å	7 µm	**711570.10**	**711570.100**
POLYGOSIL® 100-10 C_{18}	yes	14 % C	100 Å	10 µm	**711580.10**	**711580.100**
POLYGOSIL® 300-7 C_{18}	yes	4 % C	300 Å	7 µm	**711710.10**	**711710.100**
POLYGOSIL® 1000-7 C_{18}	yes	~ 1 % C	1000 Å	7 µm	**711992.10**	**711992.100**
Octyl phases						**$-(CH_2)_7-CH_3$**
POLYGOSIL® 60-5 C_8	no	7 % C	60 Å	5 µm	**711300.10**	**711300.100**
POLYGOSIL® 60-7 C_8	no	7 % C	60 Å	7 µm	**711310.10**	**711310.100**
POLYGOSIL® 60-10 C_8	no	7 % C	60 Å	10 µm	**711320.10**	**711320.100**
Butyl phases						**$-(CH_2)_3-CH_3$**
POLYGOSIL® 300-7 C_4	yes	~ 1 % C	300 Å	7 µm	**711680.10**	**711680.100**
POLYGOSIL® 1000-7 C_4	yes	< 1 % C	1000 Å	7 µm	**711991.10**	**711991.100**
Cyano phases (nitrile)						**$-(CH_2)_3-CN$**
POLYGOSIL® 60-5 CN		~ 5 % C	60 Å	5 µm	**711380.10**	**711380.100**
POLYGOSIL® 60-10 CN		~ 5 % C	60 Å	10 µm	**711390.10**	**711390.100**
Nitro phases						**$-(CH_2)_3-C_6H_4-NO_2$**
POLYGOSIL® 60-5 NO_2		~ 4.5 % C	60 Å	5 µm	**711400.10**	**711400.100**
POLYGOSIL® 60-10 NO_2		~ 4.5 % C	60 Å	10 µm	**711410.10**	**711410.100**
Unmodified silica						**SiOH**
POLYGOSIL® 60-5			60 Å	5 µm	**711010.10**	**711010.100**
POLYGOSIL® 60-7			60 Å	7 µm	**711280.10**	**711280.100**
POLYGOSIL® 60-10			60 Å	10 µm	**711020.10**	**711020.100**
POLYGOSIL® 100-5			100 Å	5 µm	**711510.10**	**711510.100**
POLYGOSIL® 100-7			100 Å	7 µm	**711520.10**	**711520.100**
POLYGOSIL® 100-10			100 Å	10 µm	**711530.10**	**711530.100**
POLYGOSIL® 300-7			300 Å	7 µm	**711600.10**	**711600.100**
POLYGOSIL® 1000-7			1000 Å	7 µm	**711890.10**	**711890.100**
Amino phases						**$-(CH_2)_3-NH_2$**
POLYGOSIL® 60-5 NH_2		~ 3 % C	60 Å	5 µm	**711360.10**	**711360.100**
POLYGOSIL® 60-10 NH_2		~ 3 % C	60 Å	10 µm	**711370.10**	**711370.100**
Dimethylamino phases						**$-(CH_2)_3-N(CH_3)_2$**
POLYGOSIL® 60-5 $N(CH_3)_2$		~ 3.5 % C	60 Å	5 µm	**711420.10**	**711420.100**
POLYGOSIL® 60-10 $N(CH_3)_2$		~ 3.5 % C	60 Å	10 µm	**711430.10**	**711430.100**

POLYGOPREP bulk packings

- irregular silica for preparative applications
- pH stability 2 – 8

Physical properties of unmodified POLYGOPREP materials

Phase	Pore size	Pore volume	Surface (BET)	Density	Pressure stability
POLYGOPREP 60	60 Å	0.75 ml/g	350 m^2/g	0.45 g/ml	600 bar
POLYGOPREP 100	100 Å	1 ml/g	280 m^2/g	0.35 g/ml	400 bar
POLYGOPREP 300	300 Å	0.8 ml/g	100 m^2/g	0.45 g/ml	400 bar
POLYGOPREP 1000	1000 Å	0.8 ml/g	35 m^2/g	0.45 g/ml	300 bar

Modification of POLYGOPREP follows the same processes as for NUCLEOSIL® silica.

Ordering information

Phase	Endcapped	Carbon content	Pore size	Particle size	Pack of 100 g	Pack of 1 kg
Octadecyl phases						$-(CH_2)_{17}-CH_3$
POLYGOPREP 60-12 C_{18}	no*	12 % C	60 Å	10 – 15 µm	**711009.100**	**711009.1000**
POLYGOPREP 60-20 C_{18}	no*	12 % C	60 Å	15 – 25 µm	**711031.100**	**711031.1000**
POLYGOPREP 60-30 C_{18}	no*	12 % C	60 Å	25 – 40 µm	**711480.100**	**711480.1000**
POLYGOPREP 60-50 C_{18}	no*	12 % C	60 Å	40 – 63 µm	**711500.100**	**711500.1000**
POLYGOPREP 60-80 C_{18}	no*	12 % C	60 Å	63 – 100 µm	**711011.100**	**711011.1000**
POLYGOPREP 60-130 C_{18}	no*	12 % C	60 Å	63 – 200 µm	**711590.100**	**711590.1000**
POLYGOPREP 100-12 C_{18}	no*	14 % C	100 Å	10 – 15 µm	**711018.100**	**711018.1000**
POLYGOPREP 100-20 C_{18}	no*	14 % C	100 Å	15 – 25 µm	**711019.100**	**711019.1000**
POLYGOPREP 100-30 C_{18}	no*	14 % C	100 Å	25 – 40 µm	**711032.100**	**711032.1000**
POLYGOPREP 100-50 C_{18}	no*	14 % C	100 Å	40 – 63 µm	**711021.100**	**711021.1000**
POLYGOPREP 300-12 C_{18}	yes	4 % C	300 Å	10 – 15 µm	**711024.100**	**711024.1000**
POLYGOPREP 300-20 C_{18}	yes	4 % C	300 Å	15 – 25 µm	**711025.100**	**711025.1000**
POLYGOPREP 300-30 C_{18}	yes	4 % C	300 Å	25 – 40 µm	**711720.100**	**711720.1000**
POLYGOPREP 300-50 C_{18}	yes	4 % C	300 Å	40 – 63 µm	**711730.100**	**711730.1000**
POLYGOPREP 1000-30 C_{18}	yes	~ 1 % C	1000 Å	25 – 40 µm	**711028.100**	**711028.1000**
POLYGOPREP 1000-50 C_{18}	yes	~ 1 % C	1000 Å	40 – 63 µm	**711029.100**	**711029.1000**
Octyl phases						$-(CH_2)_7-CH_3$
POLYGOPREP 60-12 C_8	no*	7 % C	60 Å	10 – 15 µm	**711007.100**	**711007.1000**
POLYGOPREP 60-20 C_8	no*	7 % C	60 Å	15 – 25 µm	**711008.100**	**711008.1000**
POLYGOPREP 60-30 C_8	no*	7 % C	60 Å	25 – 40 µm	**711470.100**	**711470.1000**
POLYGOPREP 60-50 C_8	no*	7 % C	60 Å	40 – 63 µm	**711490.100**	**711490.1000**
Butyl phases						$-(CH_2)_3-CH_3$
POLYGOPREP 300-12 C_4	yes	~ 1 % C	300 Å	10 – 15 µm	**711022.100**	**711022.1000**
POLYGOPREP 300-20 C_4	yes	~ 1 % C	300 Å	15 – 25 µm	**711023.100**	**711023.1000**
POLYGOPREP 300-30 C_4	yes	~ 1 % C	300 Å	25 – 40 µm	**711690.100**	**711690.1000**
POLYGOPREP 300-50 C_4	yes	~ 1 % C	300 Å	40 – 63 µm	**711700.100**	**711700.1000**
POLYGOPREP 1000-30 C_4	yes	< 1 % C	1000 Å	25 – 40 µm	**711026.100**	**711026.1000**
POLYGOPREP 1000-50 C_4	yes	< 1 % C	1000 Å	40 – 63 µm	**711027.100**	**711027.1000**

* On request, these POLYGOPREP RP phases can be endcapped at surcharge.

POLYGOPREP irregular silica for HPLC

Phase	Endcapped	Carbon content	Pore size	Particle size	Pack of 100 g	Pack of 1 kg
Cyano phases (nitrile)						**$-(CH_2)_3-CN$**
POLYGOPREP 60-12 CN		~ 4.5 % C	60 Å	10 – 15 µm	**711015.100**	**711015.1000**
POLYGOPREP 60-20 CN		~ 4.5 % C	60 Å	15 – 25 µm	**711016.100**	**711016.1000**
POLYGOPREP 60-30 CN		~ 4.5 % C	60 Å	25 – 40 µm	**711017.100**	**711017.1000**
Amino phases						**$-(CH_2)_3-NH_2$**
POLYGOPREP 60-12 NH_2		~ 3 % C	60 Å	10 – 15 µm	**711012.100**	**711012.1000**
POLYGOPREP 60-20 NH_2		~ 3 % C	60 Å	15 – 25 µm	**711013.100**	**711013.1000**
POLYGOPREP 60-30 NH_2		~ 3 % C	60 Å	25 – 40 µm	**711014.100**	**711014.1000**

Phase	Pore size	Particle size	Pack of 100 g	Pack of 1 kg	Pack of 5 kg
Unmodified POLYGOPREP silica					**SiOH**
POLYGOPREP 60-12	60 Å	10 – 15 µm		**711001.1000**	**711001.5000**
POLYGOPREP 60-20	60 Å	15 – 25 µm		**711240.1000**	**711240.5000**
POLYGOPREP 60-30	60 Å	25 – 40 µm		**711250.1000**	**711250.5000**
POLYGOPREP 60-50	60 Å	40 – 63 µm		**711260.1000**	**711260.5000**
POLYGOPREP 60-80	60 Å	63 – 100 µm		**711270.1000**	**711270.5000**
POLYGOPREP 60-130	60 Å	63 – 200 µm		**711037.1000**	**711037.5000**
POLYGOPREP 100-12	100 Å	10 – 15 µm		**711002.1000**	**711002.5000**
POLYGOPREP 100-20	100 Å	15 – 25 µm		**711003.1000**	**711003.5000**
POLYGOPREP 100-30	100 Å	25 – 40 µm		**711540.1000**	**711540.5000**
POLYGOPREP 100-50	100 Å	40 – 63 µm		**711550.1000**	**711550.5000**
POLYGOPREP 100-80	100 Å	63 – 100 µm		**711033.1000**	**711033.5000**
POLYGOPREP 100-130	100 Å	63 – 200 µm		**711034.1000**	**711034.5000**
POLYGOPREP 300-12	300 Å	10 – 15 µm	**711004.100**	**711004.1000**	
POLYGOPREP 300-20	300 Å	15 – 25 µm	**711610.100**	**711610.1000**	
POLYGOPREP 300-30	300 Å	25 – 40 µm	**711620.100**	**711620.1000**	
POLYGOPREP 300-50	300 Å	40 – 63 µm	**711630.100**	**711630.1000**	
POLYGOPREP 1000-12	1000 Å	10 – 15 µm	**711035.100**	**711035.1000**	
POLYGOPREP 1000-20	1000 Å	15 – 25 µm	**711036.100**	**711036.1000**	
POLYGOPREP 1000-30	1000 Å	25 – 40 µm	**711005.100**	**711005.1000**	
POLYGOPREP 1000-50	1000 Å	40 – 63 µm	**711006.100**	**711006.1000**	

Silica adsorbents for low pressure column chromatography

- silica 60, pore size ~ 60 Å; pore volume ~ 0.75 ml/g; spec. surface BET ~ 500 m^2/g
  highly porous, amorphous silicic acid in the form of hard, opalescent particles, prepared by precipitation of water glass with sulphuric acid
- For higher demands on the performance of column packings we recommend our high-purity irregular POLYGOPREP silicas (see previous page).
- silica FIA for the fluorescence indicator adsorption procedure for the determination of hydrocarbon groups in the testing of liquid fuels in accordance with DIN 51791 and ASTM D 1319-58T
- The FIA method determines saturated hydrocarbons, olefins and aromatic hydrocarbons of a sample chromatographically by adsorption and desorption in a column filled with FIA silica, in the presence of a fluorescent dye mixture.

Ordering information

Designation	Particle size	1 kg	5 kg	25 kg
Silica 60, 0.015 – 0.04 mm	–	**815650.1**	**815650.5**	**815650.25**
Silica 60, 0.025 – 0.04 mm	–	**815300.1**	**815300.5**	**815300.25**
Silica 60, 0.04 – 0.063 mm	230 – 400 mesh	**815380.1**	**815380.5**	**815380.25**
Silica 60 M, 0.04 – 0.063 mm	230 – 400 mesh	**815381.1**	**815381.5**	**815381.25**
Silica 60, 0.05 – 0.1 mm	130 – 270 mesh	**815390.1**	**815390.5**	**815390.25**
Silica 60, 0.05 – 0.2 mm	70 – 270 mesh	**815320.1**	**815320.5**	**815320.25**
Silica 60, 0.063 – 0.2 mm	70 – 230 mesh	**815330.1**	**815330.5**	**815330.25**
Silica 60, < 0.063 mm	+230 mesh	**815400.1**	**815400.5**	**815400.25**
Silica 60, < 0.08 mm	+190 mesh	**815310.1**	**815310.5**	**815310.25**
Silica 60, 0.1 – 0.2 mm	70 – 130 mesh	**815340.1**	**815340.5**	
Silica 60, 0.2 – 0.5 mm	35 – 70 mesh	**815350.1**	**815350.5**	**815350.25**
Silica 60, 0.5 – 1.0 mm	18 – 35 mesh	**815360.1**	**815360.5**	**815360.25**
Silica FIA fine	0.071 – 0.16 mm	**815410.1**		
Silica FIA coarse	0.071 – 0.63 mm	**815430.1**		

Aluminium oxide

- aluminium oxides produced by dehydration of different aluminium hydroxides, e.g. hydrargillite between 400 and 500 °C
- activity grade I, particle size 50 – 200 µm, specific surface (BET) ~ 130 m^2/g

Ordering information

Type	pH	1 kg	5 kg	25 kg
Aluminium oxide 90 basic	pH 9.5 ± 0.3	**815010.1**	**815010.5**	**815010.25**
Aluminium oxide 90 neutral	pH 7 ± 0.5	**815020.1**	**815020.5**	**815020.25**
Aluminium oxide 90 acidic	pH 4 ± 0.3	**815030.1**	**815030.5**	**815030.25**

Kieselguhr

- naturally occurring amorphous silicic acids of fossil origin, also known as diatomaceous earth or diatomite purified for chromatographic applications
- compared to silica, kieselguhr has a small surface of low activity → application in partition chromatography; impregnated with various substances (paraffin, silicone oil, undecane) it can be used for reversed phase chromatography
- The following grades of kieselguhr are manufactured by Johns-Manville. They are narrowly classified with homogeneous particle size distributions and high purity.

For columns packed with kieselguhr please see CHROMABOND® XTR for liquid-liquid extraction, page 54.

Ordering information

Designation	rel. purification factor	rel. flow rate	1 kg	5 kg
Filter-Cel	100	100	**815510.1**	**815510.5**
Standard Super-Cel	85	213	**815520.1**	**815520.5**
Hyflo Super-Cel	58	534	**815530.1**	**815530.5**
Celite 503	42	910	**815540.1**	**815540.5**
Celite 535	35	1269	**815550.1**	**815550.5**
Celite 545	32	1830	**815560.1**	**815560.5**

Adsorbents for column chromatography

Florisil®

- hard granular magnesia silica gel: MgO 15.5 ± 0.5 % · SiO_2 84.0 ± 0.5 % · Na_2SO_4 ≤ 1.0 %; 60/100 mesh
  typical applications: sample preparation (see chapter "Solid phase extraction", page 32); clean-up of pesticide residues, separation of chlorinated pesticides, extraction of steroids, sex hormones, antibiotics, lipids etc.

Ordering information

Designation	Particle size	1 kg	5 kg
Florisil standard 60/100 mesh	0.15/0.25 mm	**815710.1**	**815710.5**

Polyamide

- polyamide 6 = ε-aminopolycaprolactam
  separation mechanism mainly based on hydrogen bonds
  recommended application: separation of phenolic compounds (e. g. isolation of natural products), carboxylic acids, aromatic nitro compounds

For SPE columns packed with polyamide see CHROMABOND® PA page 32.

Ordering information

Designation	Particle size	1 kg	5 kg
Polyamide CC 6, < 0.07 mm	< 0.07 mm	**815610.1**	
Polyamide CC 6, 0.05 – 0.16 mm	0.05 – 0.16 mm	**815620.1**	**815620.5**
Polyamide CC 6, 0.10 – 0.30 mm	0.10 – 0.30 mm	**815600.1**	**815600.5**

Unmodified cellulose

- **cellulose MN 100:** native fibrous cellulose, standard grade
  average degree of polymerisation 620 – 680, fibre length (85 %) 20 – 100 µm,
  specific surface acc. to Blaine ~ 6500 cm^2/g
  residue on ignition at 850 °C < 10000 ppm, < 20 ppm Fe, < 5 ppm Cu, < 7 ppm P, CH_2Cl_2 extract < 0.20 %
- **cellulose MN 2100:** native fibrous cellulose, purified grade (washed with different eluents)
  average degree of polymerisation 620 – 680, fibre length (85 %) 20 – 75 µm,
  specific surface acc. to Blaine ~ 5500 cm^2/g
  residue on ignition at 850 °C < 1000 ppm, < 2 ppm Fe, < 1 ppm Cu, < 2 ppm P, CH_2Cl_2 extract < 0.15 %
  grade MN 2100ff is a defatted cellulose MN 2100 with a CH_2Cl_2 extract < 0.02 %

Ordering information

Designation	1 kg	5 kg	25 kg
Cellulose MN 100	**815050.1**	**815050.5**	**815050.25**
Cellulose MN 2100	**815060.1**	**815060.5**	**815060.25**
Cellulose MN 2100ff (cellulose MN 2100 defatted)	**815070.1**		