

Chromatography

# NUCLEODUR®

## Professional solutions for HPLC



An optimized phase for every field of application



**MACHEREY-NAGEL**

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## Meeting your needs

If you have any questions concerning our NUCLEODUR® program or other chromatography products, please feel free to contact us:

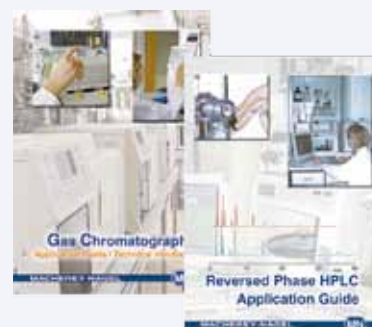
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## Ask for our application guides



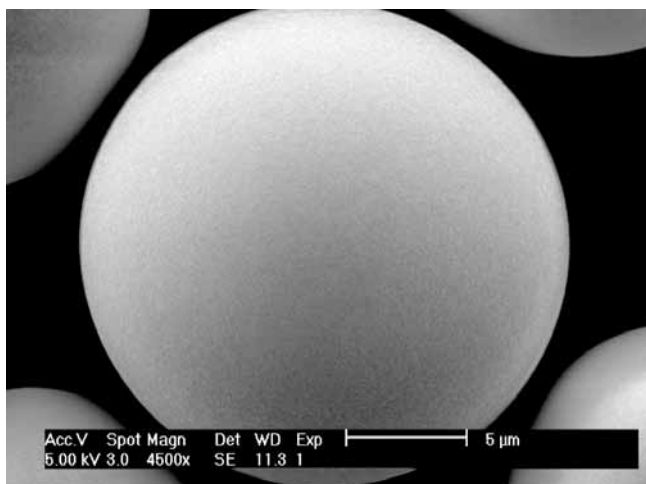
## NUCLEODUR®

NUCLEODUR® is a fully synthetic type B silica (silica of 3<sup>rd</sup> generation) offering highly advanced physical properties like **totally spherical** particle shape, outstanding **surface smoothness**, 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 worthy successor of MN's world 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 (see appl. 118630 on page 63).

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 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 <sup>2</sup> /g
Pore size	110 Å
Pore volume	0.9 mL/g



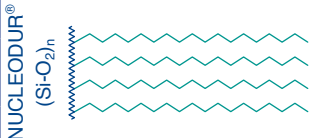




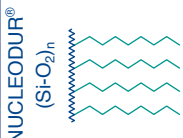




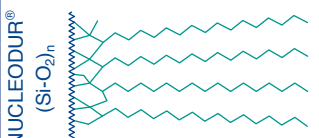




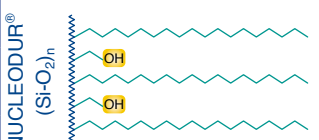




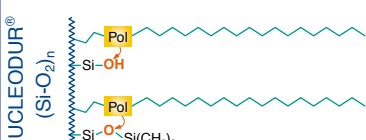




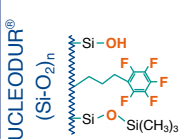




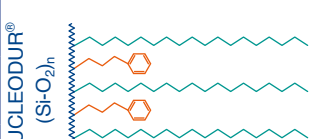




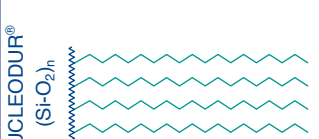


### NUCLEODUR® modifications

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<sub>18</sub> Gravity and C<sub>8</sub> Gravity
- NUCLEODUR® C<sub>18</sub> Isis
- NUCLEODUR® C<sub>18</sub> Pyramid
- NUCLEODUR® PolarTec
- NUCLEODUR® PFP
- NUCLEODUR® Sphinx RP
- NUCLEODUR® C<sub>18</sub> HTec
- NUCLEODUR® C<sub>18</sub> ec and C<sub>8</sub> ec
- NUCLEODUR® HILIC
- NUCLEODUR® CN and CN-RP
- NUCLEODUR® NH<sub>2</sub> and NH<sub>2</sub>-RP
- unmodified NUCLEODUR®

All phases are described in detail on the following pages.

# Overview of NUCLEODUR® HPLC phases

Phase	Specification	Characteristics*	Stability	Structure
	octadecyl phase, high density coating, multi-encapping 18% C · USP L1	A 	pH stability 1-11, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
		B 		
		C 		
	octyl phase, high density coating, multi-encapping 11% C · USP L7	A 	pH stability 1-11, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
		B 		
		C 		
	octadecyl phase with specially crosslinked surface modification, endcapping 20% C · USP L1	A 	pH stability 1-10, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
		B 		
		C 		
	C <sub>18</sub> modification with polar endcapping 14% C · USP L1	A 	stable against 100% aqueous eluents, pH stability 1-9, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
		B 		
		C 		
	octadecyl phase with embedded polar group, endcapping 15.5% C · USP L1 and L60	A 	stable against 100% aqueous eluents, pH stability 1-9, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
		B 		
		C 		
	pentafluorophenyl-propyl modification with multi- endcapping 8% C · USP L43	A 	pH stability 1-9, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
		B 		
		C 		
	bifunctional RP phase, propylphenyl and C <sub>18</sub> li- gands, endcapping 15% C · USP L1 and L11	A 	pH stability 1-10, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
		B 		
		C 		
	octadecyl phase with high capacity, high density coat- ing, multi-encapping 18% C · USP L1	A 	pH stability 1-11, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
		B 		
		C 		

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


# An optimized phase for every separation

Application	Similar phases**	Separation principle · Retention mechanism	
in general compounds with ionizable functional groups such as basic pharmaceuticals and pesticides	<b>NUCLEOSIL® C<sub>18</sub> HD</b> XTerra® RP18 / MS C <sub>18</sub> ; Luna® C18(2), Gemini®, Synergi® Max RP; Zorbax® Extend-C18; Inertsil® ODS III; Purospher® STAR RP-18; Hypersil™ BDS	<b>only hydrophobic interactions</b> (van der Waals interactions)	
like C <sub>18</sub> Gravity, however generally shorter retention times for nonpolar compounds	<b>NUCLEOSIL® C<sub>8</sub> HD</b> XTerra® RP8 / MS C <sub>8</sub> ; Luna® C8; Zorbax® Eclipse XDB-C8	<b>only hydrophobic interactions</b> (van der Waals interactions)	
high steric selectivity, thus suited for separation of positional and structural isomers, planar/nonplanar molecules	<b>NUCLEOSIL® C<sub>18</sub> AB</b> Inertsil® ODS-P; Pro C18 RS; Zorbax® SB	<b>steric interactions and hydrophobic interactions</b>	
basic pharmaceutical ingredients, very polar compounds, organic acids	Aqua, Synergi® Hydro-RP; AQ; Atlantis® dC <sub>18</sub>	<b>hydrophobic interactions and polar interactions</b> (H bonds)	
basic pharmaceuticals, organic acids, pesticides, amino acids, water-soluble vitamins	<b>NUCLEOSIL® C<sub>18</sub> Nautilus</b> ProntoSIL® C18; Zorbax® Bonus-RP, Polariss® Amide-C18; Ascentis® RP Amide; SymmetryShield™ RP18; SUPELCOSIL™ LC-ABZ+; HyPURITY™ ADVANCE	<b>hydrophobic interactions and polar interactions</b> (H bonds)	
aromatic and unsaturated compounds, halogen compounds, phenols, isomers, polar pharmaceuticals, antibiotics	ACQUITY® CSH Fluoro-Phenyl; Hypersil™ GOLD PFP; Luna® PFP(2); Discovery® HS F5; Allure® PFP Propyl, Ultra II PFP Propyl	<b>polar interactions</b> (H bonds), <b>dipole-dipole interactions</b> <b>π-π interactions</b> and <b>hydrophobic interactions</b>	
compounds with aromatic and multiple bond systems	no similar phases	<b>π-π interactions and hydrophobic interactions</b>	
robust and well base deactivated C <sub>18</sub> phase; all separation tasks with preparative potential	XTerra® RP18 / MS C <sub>18</sub> / SunFire™ C <sub>18</sub> ; Luna® C18(2), Gemini®, Synergi® Max RP; Zorbax® Extend-C18; Inertsil® ODS III; Purospher® STAR RP-18; Hypersil™ BDS	<b>only hydrophobic interactions</b> (van der Waals interactions)	

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

# Overview of NUCLEODUR® HPLC phases

Phase	Specification	Characteristics*	Stability	Structure
 <b>C<sub>18</sub> ec</b>	octadecyl phase, medium density coating endcapping 17.5 % C · USP L1	A  B  C 	pH stability 1-9	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
 <b>C<sub>8</sub> ec</b>	octyl phase, medium density coating endcapping 10.5 % C · USP L7	A  B  C 	pH stability 1-9	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
 <b>HILIC</b>	zwitterionic ammonium sulfonic acid modification 7 % C	A  B  C -	pH stability 2-8.5, suitable for LC/MS	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
 <b>CN / CN-RP</b>	cyano (nitrile) phase for NP and RP separations 7 % C · USP L10	A  B  C -	pH stability 1-8, stable towards highly aqueous mobile phases	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
 <b>NH<sub>2</sub> / NH<sub>2</sub>-RP</b>	amino phase for NP and RP separations 2.5 % C · USP L8	A  B  C -	pH stability 2-8, stable towards highly aqueous mobile phases	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 
 <b>SiOH</b>	unmodified high purity silica USP L3	A - B n.a. C -	pH stability 2-8	NUCLEODUR® (Si-O <sub>2</sub> ) <sub>n</sub> 

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



# An optimized phase for every separation

Application	Similar phases**	Separation principle · Retention mechanism
robust C <sub>18</sub> phase for routine analyses	<b>NUCLEOSIL® C<sub>18</sub></b> Spherisorb® ODS II, Symmetry® C <sub>18</sub> ; Hypersil™ ODS; Inertsil® ODS II; Kromasil C <sub>18</sub> ; LiChrospher® RP-18	<b>only hydrophobic interactions</b> (van der Waals interactions) some residual silanol interactions
robust C <sub>8</sub> phase for routine analyses	<b>NUCLEOSIL® C<sub>8</sub> ec / C<sub>8</sub></b> Spherisorb® C <sub>8</sub> , Symmetry® C <sub>8</sub> ; Hypersil™ MOS; Kromasil C <sub>8</sub> ; LiChrospher® RP-8	<b>only hydrophobic interactions</b> (van der Waals interactions) some residual silanol interactions
hydrophilic compounds such as organic polar acids and bases, polar natural compounds	SeQuant™ ZIC®-HILIC; Obelisc™	<b>ionic / hydrophilic interactions, electrostatic interactions</b>
polar organic compounds (basic drugs), molecules containing π electron systems	<b>NUCLEOSIL® CN / CN-RP</b>	<b>π-π interactions, polar interactions (H bonds), hydrophobic interactions</b>
sugars, sugar alcohols and other hydroxy compounds, DNA bases, polar compounds in general	<b>NUCLEOSIL® NH<sub>2</sub> / NH<sub>2</sub>-RP</b>	<b>polar / ionic interactions, hydrophobic interactions</b>
polar compounds in general	<b>unmodified NUCLEOSIL®</b>	<b>polar / ionic interactions</b>

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



# 1.8 µm particle size

## 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<sub>18</sub> Gravity  
C<sub>8</sub> Gravity  
C<sub>18</sub> Isis  
C<sub>18</sub> Pyramid  
Sphinx RP  
C<sub>18</sub> HTec  
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 1.8 µm NUCLEODUR® particles researchers have turned over a new leaf in HPLC column technology. Columns packed with these microspherical 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<sub>18</sub> 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 app. 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<sub>s</sub> = resolution  
α = selectivity (separation factor)  
k<sub>i</sub>' = retention  
N = plate number with N ∝ 1/d<sub>p</sub>  
d<sub>p</sub> = particle size

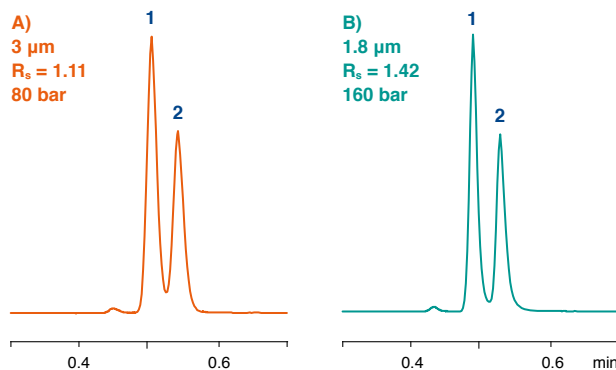
## Resolution as a function of particle size

Column: 50 x 4 mm NUCLEODUR® C<sub>18</sub> 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

A)  
3 µm  
R<sub>s</sub> = 1.11  
80 bar

B)  
1.8 µm  
R<sub>s</sub> = 1.42  
160 bar





# Increase in separation efficiency

## Column back pressure

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

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

$\Delta_P$  = pressure drop  
 $\Phi$  = flow resistance (nondimensional)  
 $L_C$  = column length  
 $\eta$  = 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 bars and the entire system should feature the lowest possible dead volume.

### Comparison of back pressures:

Eluent: 100% methanol  
 Flow rate: 1.5 mL/min  
 Temperature: 22 °C  
 Column dimension: 50 x 4.6 mm

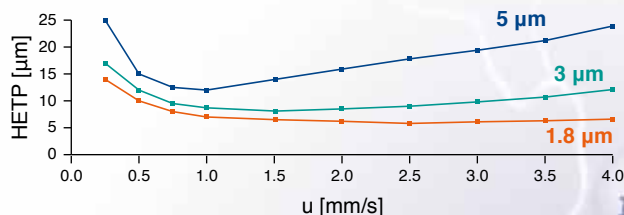
	NUCLEODUR® C <sub>18</sub> Gravity	Competitor A
3 µm	70 bar	-
1.8 µm	130 bar	170 bar

## 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



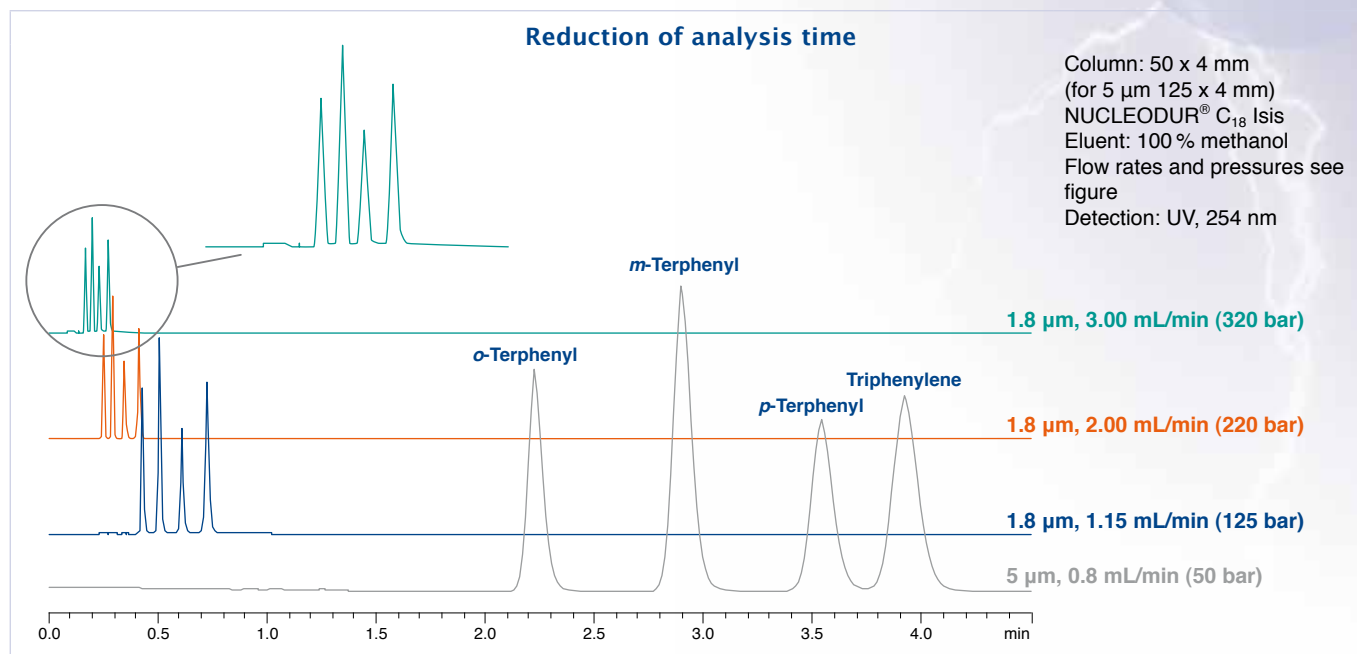
## Technical requirements

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.

Currently the following NUCLEODUR® premium phases (C<sub>18</sub> Gravity, C<sub>8</sub> Gravity, C<sub>18</sub> Isis, C<sub>18</sub> Pyramid, Sphinx RP, C<sub>18</sub> HTec, HILIC) are available in 1.8 µm. The description of each phase and its selectivity can be found in the individual chapters.

More applications on NUCLEODUR® 1.8 µm can be found in the "Applications" section from page 32.

## Reduction of analysis time



# C<sub>18</sub> Gravity / C<sub>8</sub> Gravity

## 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<sub>18</sub>) and octyl (C<sub>8</sub>), multi-encapped; pore size 110 Å; particle sizes 1.8 µm, 3 µm and 5 µm for C<sub>18</sub>, 1.8 and 5 µm for C<sub>8</sub>; 7, 10, 12 and 16 µm particles for preparative purposes on request; carbon content 18% for C<sub>18</sub>, 11% for C<sub>8</sub>

## Recommended application:

Overall sophisticated analytical separations

Compound classes separated include: pharmaceuticals, e.g., analgesics, anti-inflammatory drugs, antidepressants; herbicides; phytopharmaceuticals; immunosuppressants

USP L1 (C<sub>18</sub>) / USP L7 (C<sub>8</sub>)

## Base deactivation

NUCLEODUR® C<sub>18</sub> Gravity and NUCLEODUR® C<sub>8</sub> Gravity are based on the ultrapure NUCLEODUR® silica.

A unique derivatization process generates a homogeneous surface with a high density of bonded silanes (carbon content ~18% for C<sub>18</sub>, ~11% for C<sub>8</sub>). 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. The figure on the right shows a comparison study, where the strongly basic amitriptyline is eluted on various highly base deactivated C<sub>18</sub> phases under isocratic conditions. For a discussion of the different retention behavior of octadecyl phases compared to octyl phases see page 25.

## Tanaka diagrams

Several NUCLEODUR® phases have been examined in accordance with Tanaka et al. [J. Chromatogr. Sci. 27 (1989) 721] and Johnson et al. [Chromatographia 44 (1997) 151] with respect to the following parameters:

**Capacity** =  $k'$ (pentylbenzene)

**Hydrophobicity** =  $\alpha$ (pentylbenzene, butylbenzene)

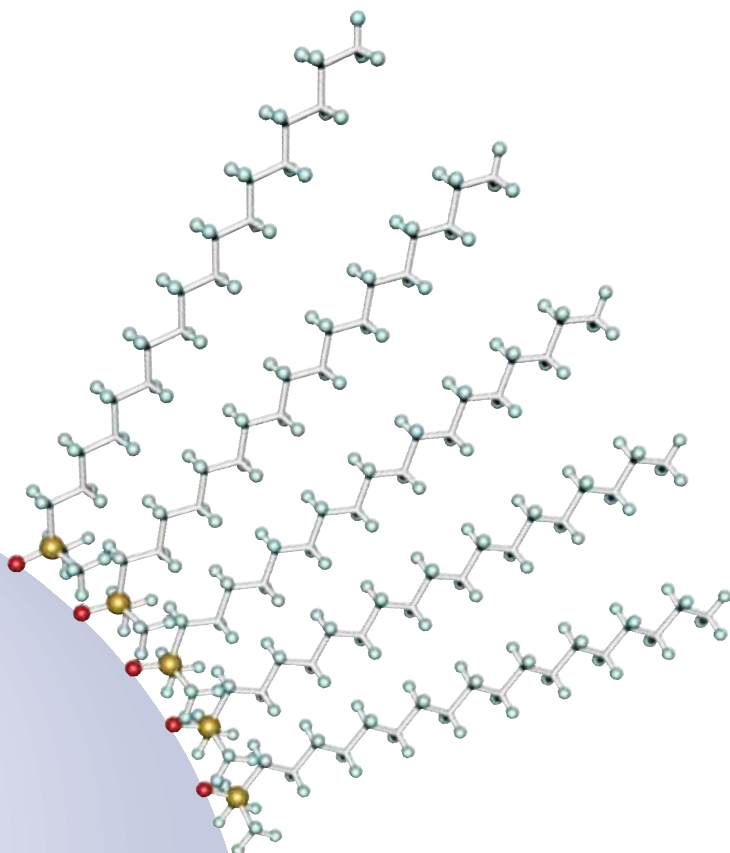
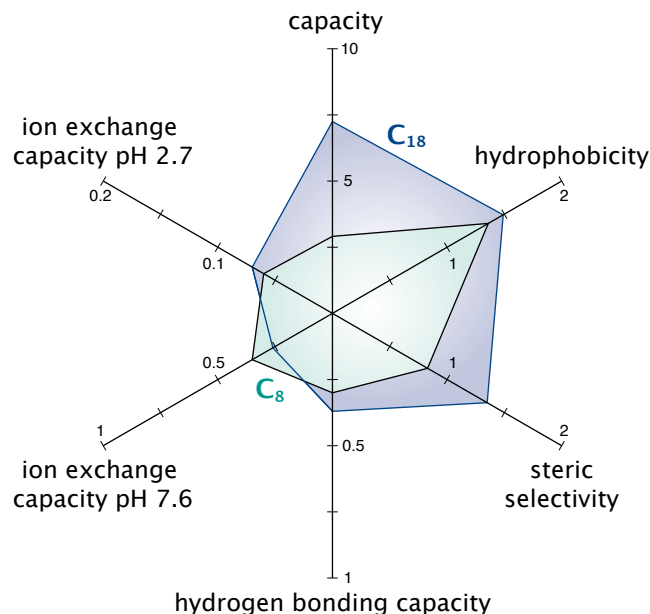
**Steric selectivity** =  $\alpha$ (triphenylene, o-terphenyl)

**Hydrogen bonding capacity (silanol capacity)** =  $\alpha$ (caffeine, phenol)

**Ion exchange capacity** at 2 different pH values (2.7 and 7.6) =  $\alpha$ (benzylamine, phenol)

The resulting Tanaka plots are shown with the respective phases.

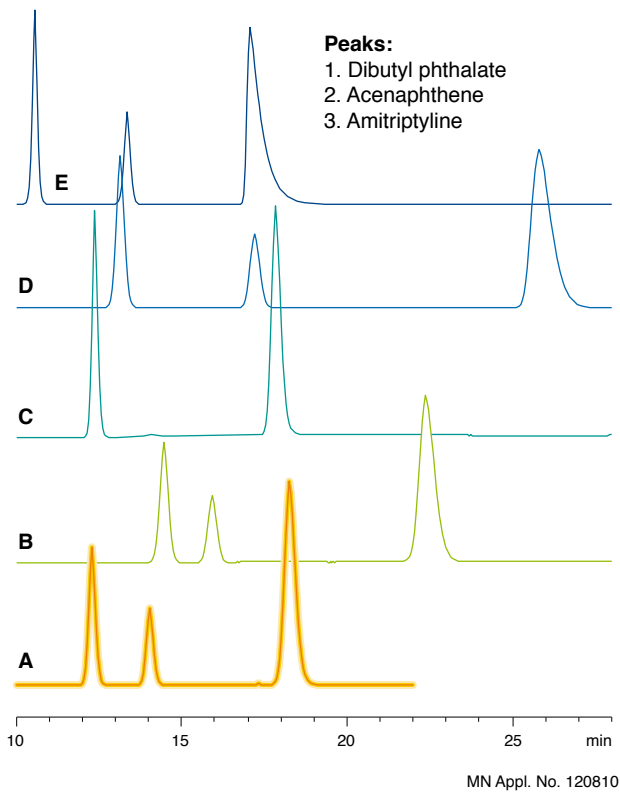
## Tanaka plots of NUCLEODUR® C<sub>8</sub> and C<sub>18</sub> Gravity



## Comparison of different base deactivated phases

Columns: 250 x 4 mm, all phases C<sub>18</sub>, 5 μm  
 A) NUCLEODUR Gravity  
 B) phase I  
 C) phase L (1 and 2 overlap)  
 D) phase P  
 E) phase S

Eluent: methanol – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 7.0 (75:25, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 30 °C  
 Detection: UV, 254 nm



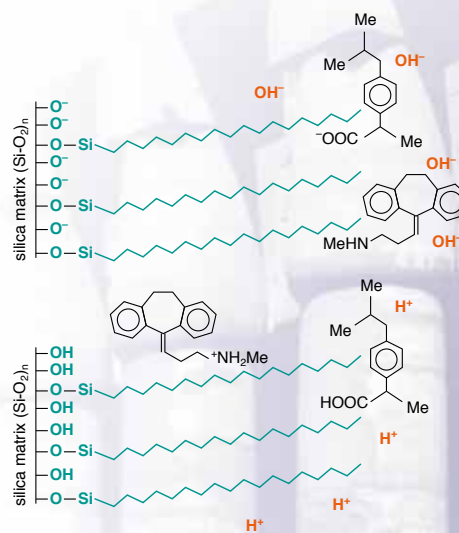
## Enhanced pH stability

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<sub>8</sub> / C<sub>18</sub> Gravity allow for use at an expanded pH range from pH 1 to 11.

## When is enhanced pH stability beneficial?

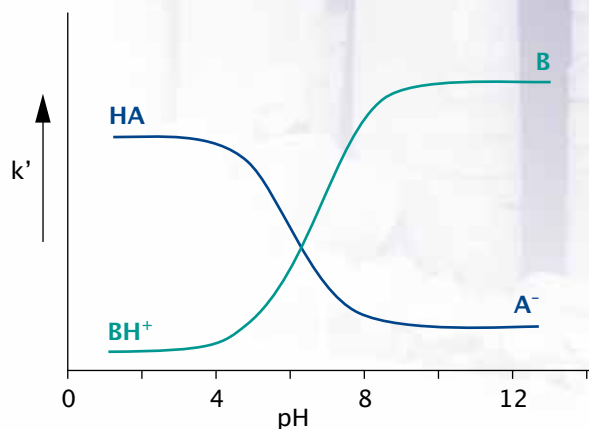
The option to work at an expanded pH range is often required in method development. Many nitrogen containing compounds like basic drugs are protonated at acidic or neutral pH and exhibit poor retention on a standard C<sub>18</sub> phase. The retention behavior can be improved by working at a higher pH, 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



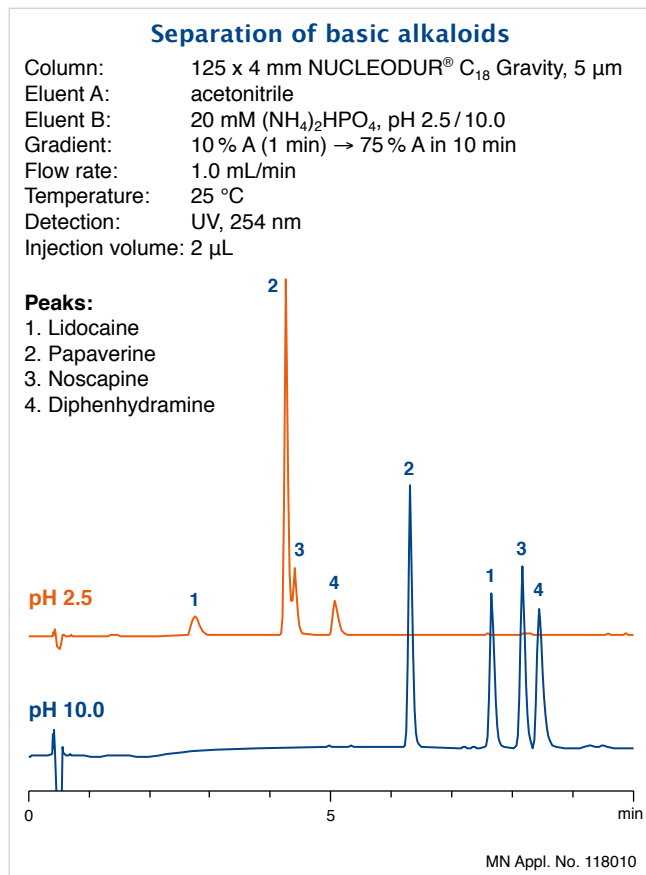
The figure above shows the extent of protonation of surface silanols and of two exemplary analytes at acidic and alkaline pH. The following graph explains the general correlation between retention and pH.

## Correlation between retention and pH for basic and acidic compounds



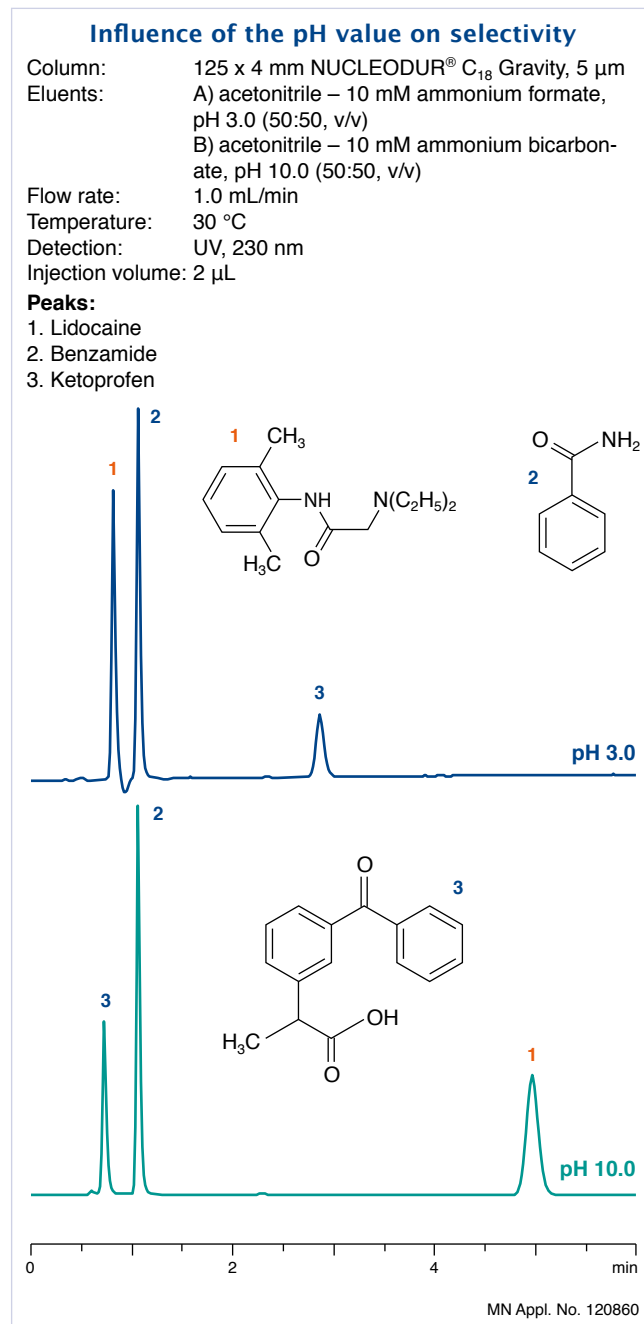
# C<sub>18</sub> Gravity / C<sub>8</sub> Gravity

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.



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<sub>18</sub> 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.



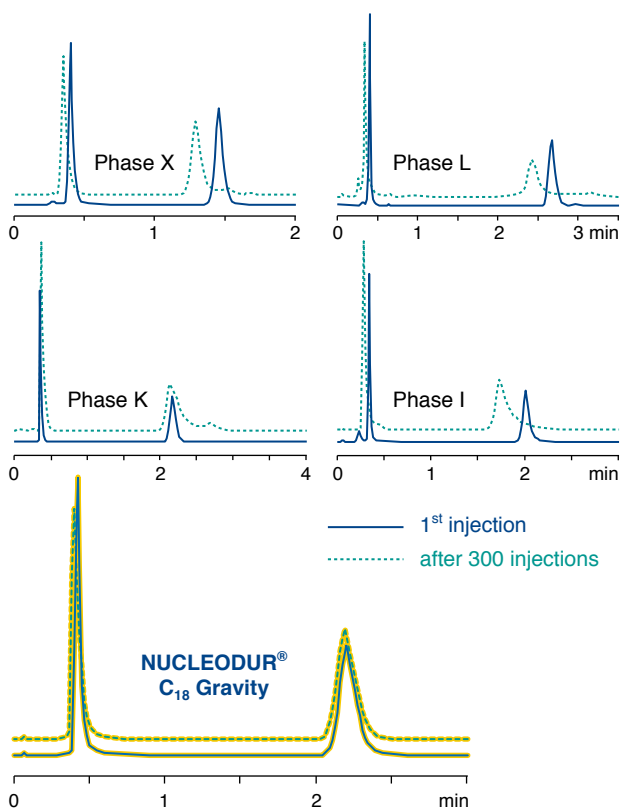
The following chromatograms demonstrate the stability of NUCLEODUR® C<sub>18</sub> Gravity under alkaline conditions in comparison with 4 commercially available modern RP18 phases. Again, the ultrapure Gravity with its unique high density surface bonding technology withstands strong alkaline mobile phase conditions. Even after 300 injections no loss of column efficiency, identified, e.g., by peak broadening or decrease in retention times, could be observed.

## Stability of NUCLEODUR® C<sub>18</sub> Gravity under alkaline conditions compared with different C<sub>18</sub> 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



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.

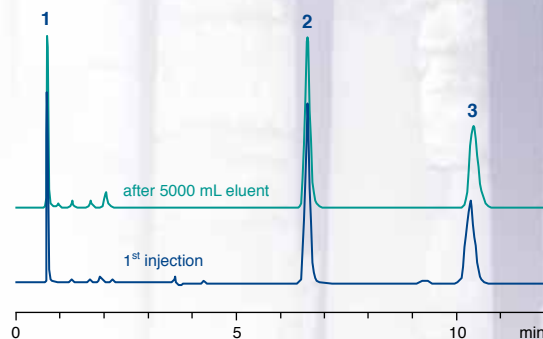
The following chromatograms show the excellent column stability of NUCLEODUR® C<sub>18</sub> Gravity in acidic conditions. The retention time of all three compounds in the column performance test remains consistent and virtually unchanged, even after the column is run with 5000 mL eluent. Due to the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

## Stability of NUCLEODUR® C<sub>18</sub> Gravity at pH 1.5

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> 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

For comparison of the selectivity of NUCLEODUR® C<sub>8</sub> Gravity and C<sub>18</sub> Gravity please also see the application "Retention behavior of different NUCLEODUR® phases" on page 15. Some general selection criteria and principals of different retention and selectivity of C<sub>18</sub> and C<sub>8</sub> columns can be found on page 25.

# C<sub>18</sub> Isis

## Key features:

- Exceptional steric selectivity
- Outstanding surface deactivation
- Suitable for LC/MS and HPLC at pH 1–10

## Technical characteristics:

C<sub>18</sub> 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

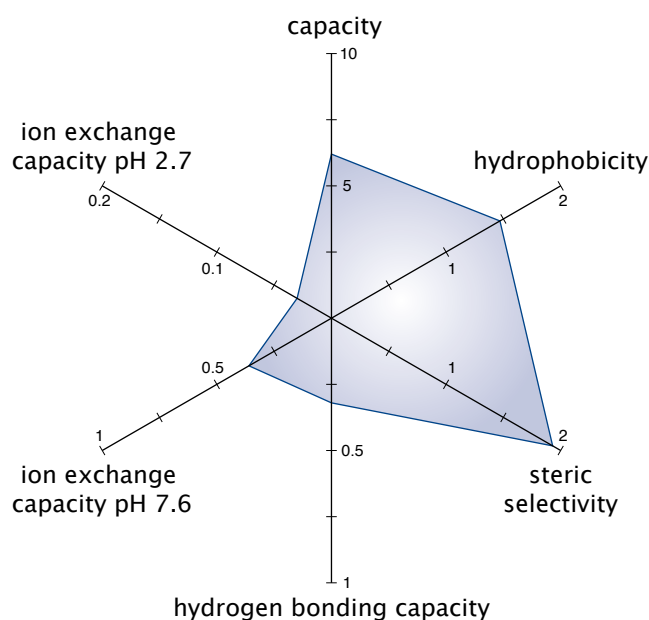
## Surface modification

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

The target crosslinking of the C<sub>18</sub> 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<sub>18</sub> Isis (1) in direct comparison with monomerically coated (2) and polar endcapped (3) C<sub>18</sub> columns.

## Tanaka plot of NUCLEODUR® C<sub>18</sub> Isis



## Steric selectivity of NUCLEODUR® C<sub>18</sub> Isis

Columns: 125 x 4 mm; NUCLEODUR® C<sub>18</sub> Isis, monomerically coated C<sub>18</sub> phase, polar endcapped phase

Eluent: methanol – water (90:10, v/v)

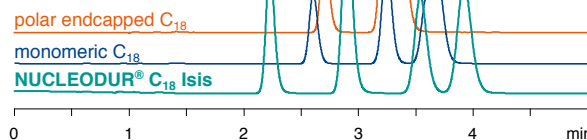
Flow rate: 1 mL/min, temperature: 35 °C

Detection: UV, 254 nm

Injection volume: 5 µL

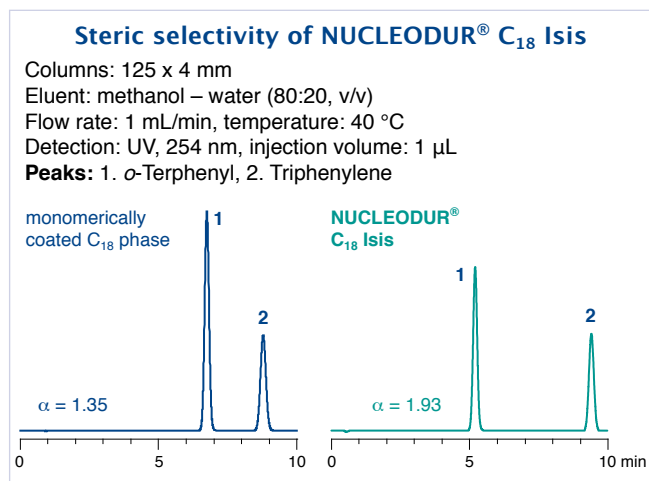
### Peaks:

1. *o*-Terphenyl
2. *m*-Terphenyl
3. *p*-Terphenyl
4. Triphenylene



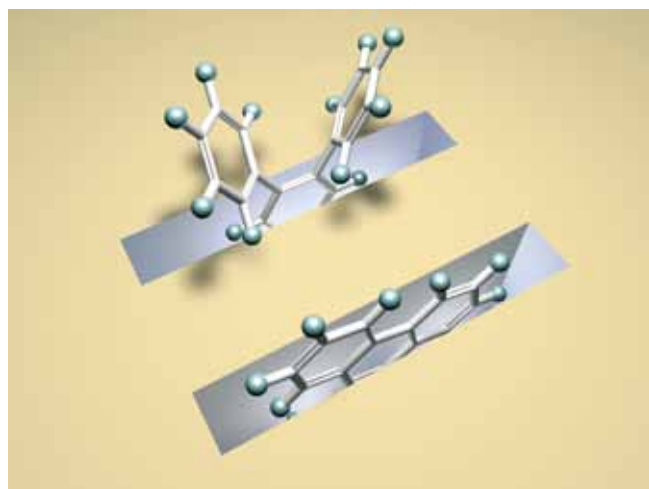
The separation of *o*-terphenyl and triphenylene is a concrete example to evaluate the selectivity potential of a reversed phase column in terms of the different shape of two molecules. The phenyl rings of *o*-terphenyl are twisted out of plane while triphenylene has a planar geometry.

The separation factor ( $\alpha$ -value) is a measure for the steric selectivity. As is shown in the following chromatograms the  $\alpha$ -value is considerable larger on NUCLEODUR® C<sub>18</sub> Isis compared to a conventional C<sub>18</sub> column.



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<sub>18</sub> 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 nonplanar molecules of similar molecular weight and length-to-breadth ratio. Thus triphenylene is longer retained than *o*-terphenyl.

## Slot model

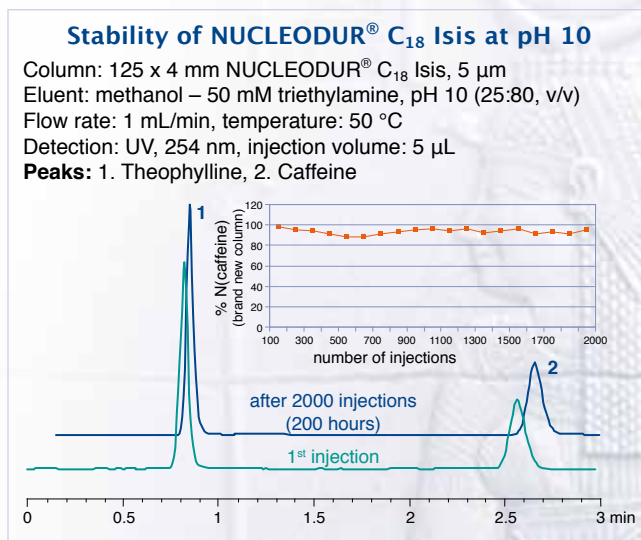


## Surface deactivation

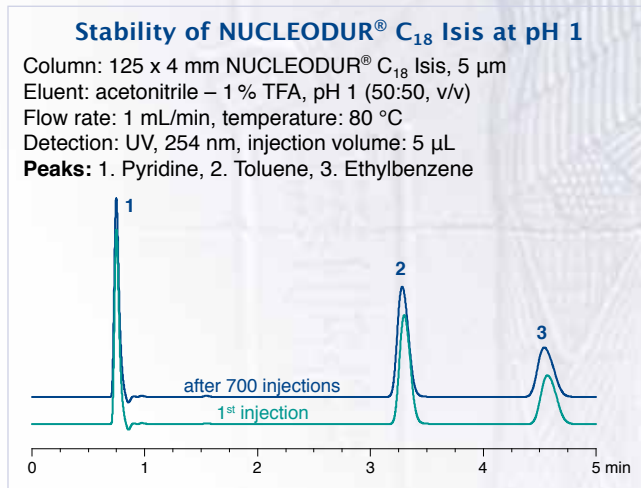
The chromatography of basic analytes requires a high density of surface-bonded C<sub>18</sub> silanes combined with a thorough endcapping procedure to keep silanol activity at a minimum. This ensures tailing-free elution of even strongly basic amino-containing compounds (see appl. 121210 on page 35).

## Stability

The applied special surface bonding technology also provides improved stability features for the NUCLEODUR® C<sub>18</sub> Isis phase. The proof for this was given in a long-term test in which the decrease of plate counts for caffeine at pH 10 and 50 °C has been observed over a period of 200 hours and 2000 sample injections, respectively. In addition retention and peak shape of caffeine and theophylline were compared with the chromatographic performance of the brand new column.



The following chromatograms exhibit the excellent stability of NUCLEODUR® C<sub>18</sub> Isis at pH 1 and 80 °C. After 700 column runs retention time and peak shape of the three test compounds remain almost unchanged.



- NUCLEODUR® C<sub>18</sub> Isis does not show any degradation under the applied mobile phase conditions. An enhanced pH stability in the range from pH 1 to 10 can be certified for this phase.

# C<sub>18</sub> Pyramid

## 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

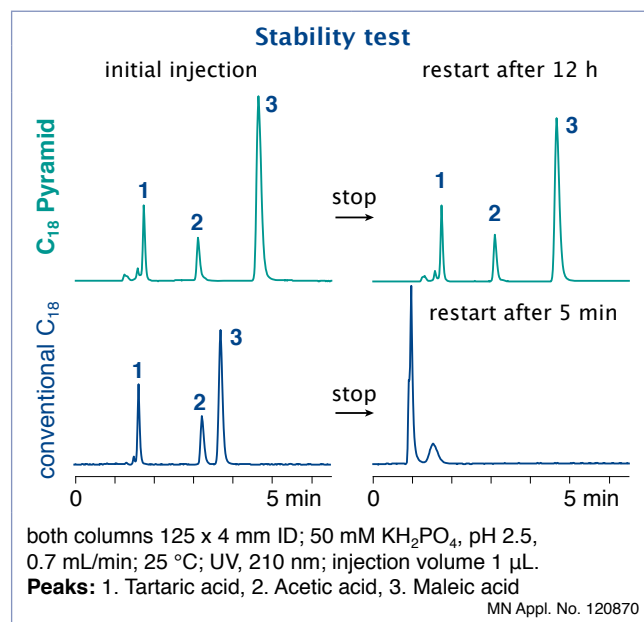
## RP-HPLC with highly aqueous mobile phases

The efforts to neutralize unwanted activity of unreacted surface silanols 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. NUCLEODUR® PolarTec may be taken as an example for the embedded polar group strategy, in which a C<sub>18</sub> silane with a polar function is successfully linked to the silica surface.

## Stability features

NUCLEODUR® C<sub>18</sub> Pyramid is a silica phase with hydrophilic endcapping, designed especially for use in eluent systems of up to 100% water. The figure below shows the retention behavior of tartaric, acetic and maleic acid under purely aqueous conditions on NUCLEODUR® C<sub>18</sub> Pyramid in comparison with a conventionally bonded RP phase.

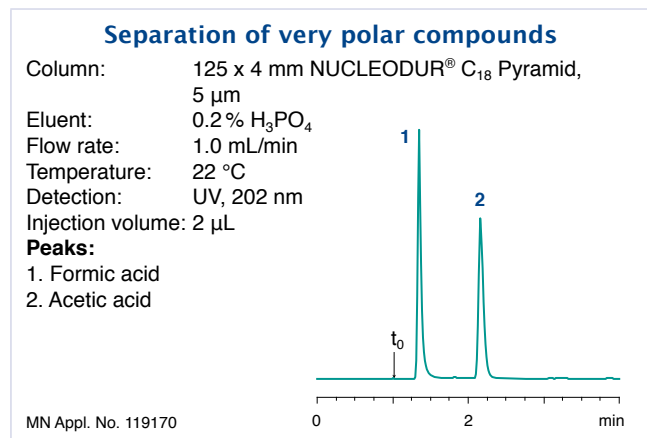


It can be shown that the retention times for NUCLEODUR® C<sub>18</sub> Pyramid remain nearly unchanged between initial injection and restart after the flow has been stopped for 12 hours, whilst the performance of the conventional RP column already collapsed totally after 5 min.



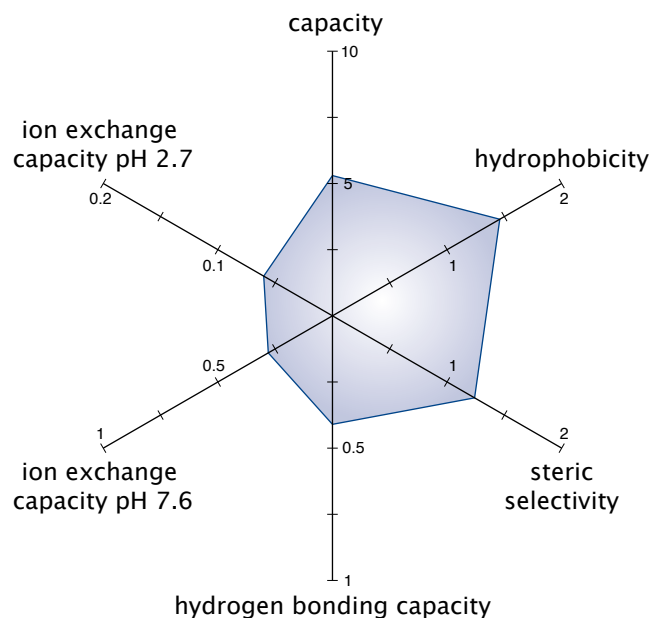
## Retention characteristics

Based on the ultrapure NUCLEODUR® silica the polar surface derivatization exhibits retention characteristics, which differentiate the "Pyramid" from conventional C<sub>18</sub> stationary phases. The chromatogram below shows the improved retention behavior of very polar compounds such as short chain organic acids, which are insufficiently retained on RP columns with predominantly hydrophobic surface properties. For more separations on NUCLEODUR® C<sub>18</sub> Pyramid see the "Applications" section from page 32.



In addition to the exceptional polar selectivity NUCLEODUR® C<sub>18</sub> Pyramid also provides adequate hydrophobic retention (see application 119190 at [www.mn-net.com](http://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<sub>18</sub> phases.

## Tanaka plot of NUCLEODUR® C<sub>18</sub> Pyramid



## Base deactivation

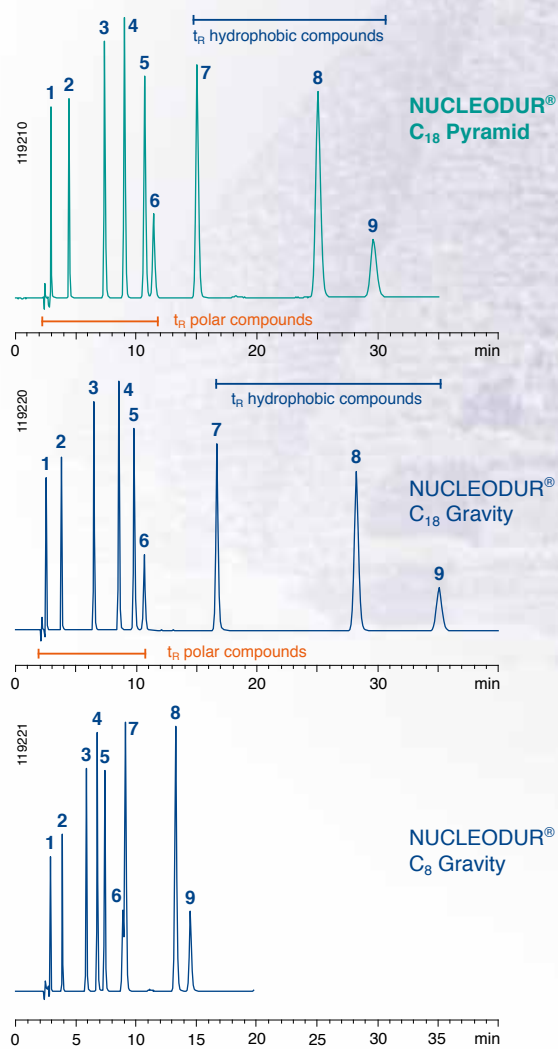
The perceptible increase in polarity has no impact on the retention behavior of ionizable analytes. Even with the strongly basic compounds of the tricyclic antidepressant drug test mixture, no unwanted interactions or a so-called lack in base deactivation are observed (see application 119200 on page 35).

## Retention behavior of polar and non-polar compounds on different NUCLEODUR® RP columns

Columns: 250 x 4 mm, 5 µm particles  
Eluent: methanol – 25 mM NH<sub>4</sub>H<sub>2</sub>PO<sub>4</sub>, pH 7 (65:35, v/v)  
Flow rate: 0.8 mL/min  
Temperature: 40 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

### Peaks:

1. Chlorpheniramine
2. Dimethyl phthalate
3. Benzamide
4. Ethyl benzoate
5. Benzophenone
6. Lidocaine
7. Naphthalene
8. Biphenyl
9. Acenaphthene



● **Key features:**

- Excellent base deactivation
- Suitable for LC/MS and stable in 100% aqueous mobile phases
- Pronounced steric selectivity

● **Technical characteristics:**

Phase with embedded polar group; pore size 110 Å; particle sizes 3 µm and 5 µm; carbon content 17%; pH stability 1–9

● **Recommended application:**

Exceptional selectivity for carbocyclic acids, phenols and nitrogen containing compounds, polar compounds like basic pharmaceuticals, organic acids, pesticides, amino acids, water soluble vitamins, etc.

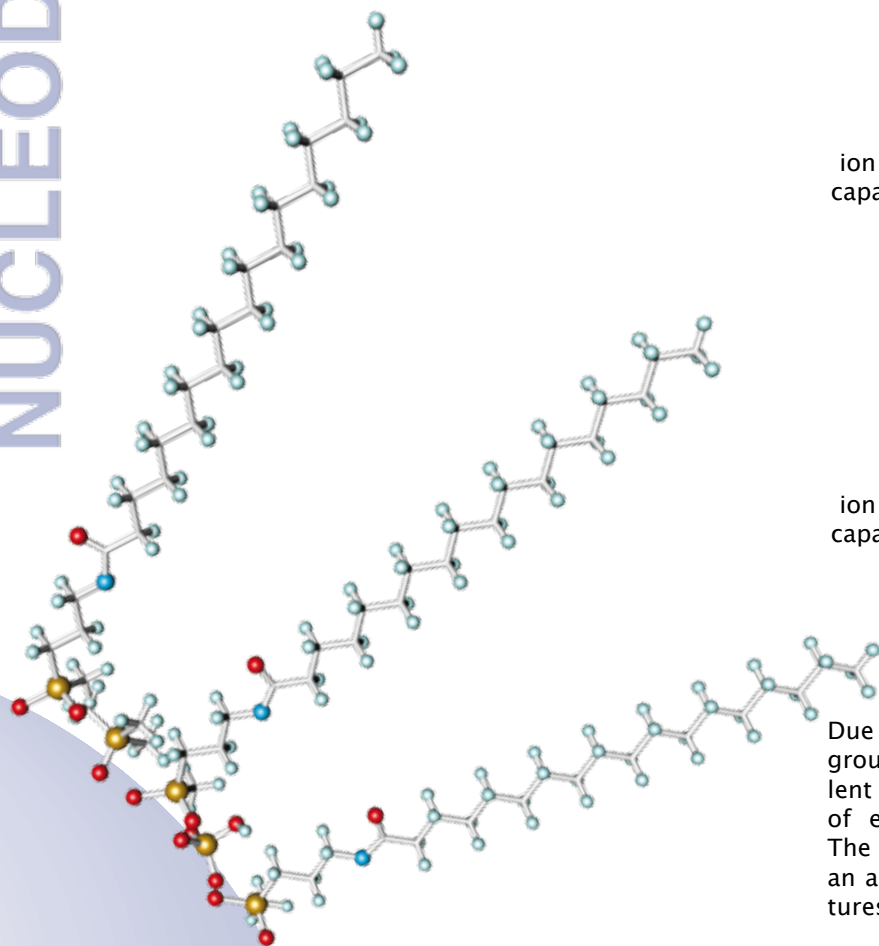
**USP L1 and L60**

## RP-HPLC under 100% aqueous conditions

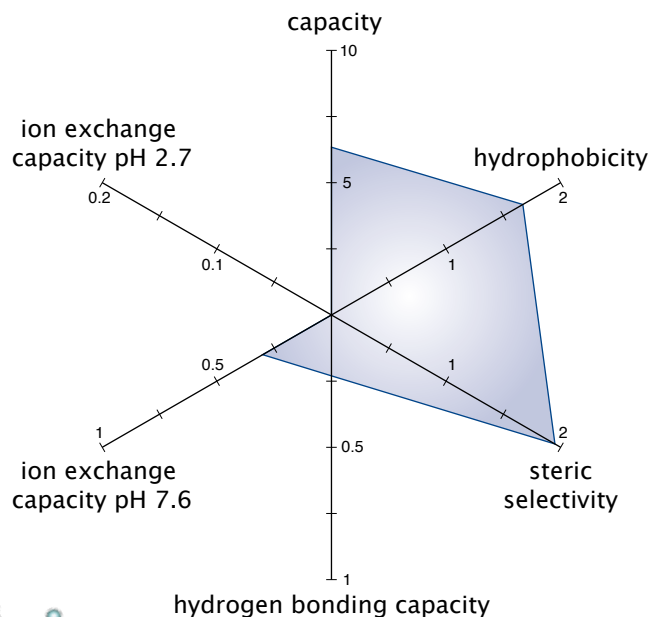
The dominant form of interactions of conventional C<sub>18</sub> phases are non-polar London dispersion forces. Besides non-polar interactions phases with embedded polar groups possess the ability to show polar interactions (dipol-dipol, hydrogen-bondings, π-π, etc.) These interactions enhance retention and selectivity for polar compounds like carbocyclic acids, phenols and nitrogen containing compounds (see applications).

In order to increase retention for polar compounds it is often necessary to decrease the organic ratio of the mobile phase to zero. Under these conditions many conventional C<sub>18</sub> phases display the so-called dewetting effect which means that the mobile phase is expelled from the pores. This phenomenon leads to a dramatic loss in retention. NUCLEODUR® PolarTec is stable in 100% aqueous mobile phases and therefore especially suited for the separation of polar compounds like organic acids (see appl. 124562 on page 55).

NUCLEODUR®



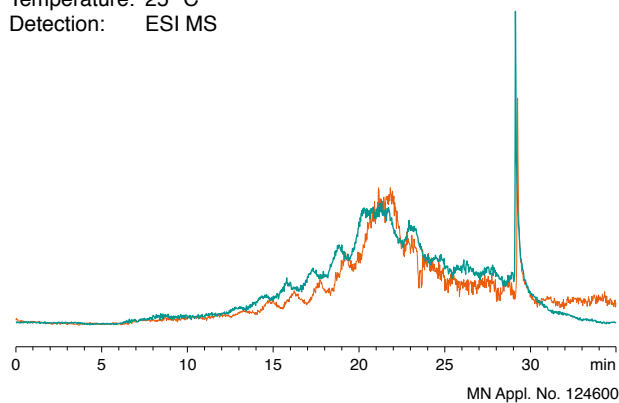
Tanaka plot of NUCLEODUR® PolarTec



Due to the shielding effect of the embedded group NUCLEODUR PolarTec shows an excellent base deactivation, which is at the top-notch of embedded polar group phases on the market. The pronounced steric selectivity (see Tanaka plot) is an additional tool for the separation of complex mixtures.

## Bleeding of NUCLEODUR® PolarTec

Columns: 150 x 3 mm NUCLEODUR® PolarTec, 5 µm  
150 x 3 mm Waters SymmetryShield™ RP18, 5 µm  
Eluent: A) acetonitrile, B) water  
Gradient: 10% B → 90% B in 10 min  
Flow rate: 0.2 mL/min  
Temperature: 25 °C  
Detection: ESI MS



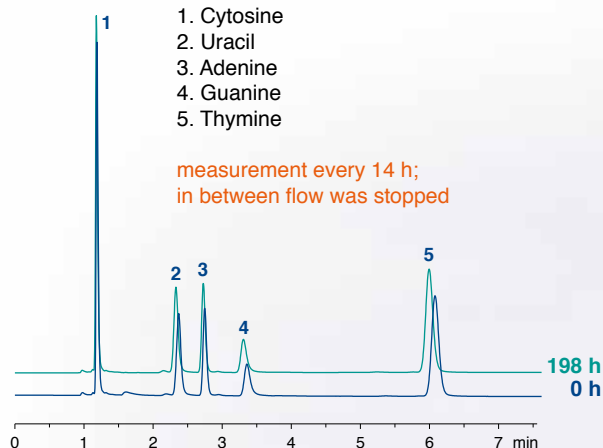
Due to low bleeding characteristics NUCLEODUR® PolarTec is also suitable for LC/MS.

## Stability of NUCLEODUR® PolarTec

Column: 150 x 3 mm NUCLEODUR® PolarTec, 3 µm  
Eluent: 30 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3.0  
Flow rate: 0.5 mL/min  
Temperature: 30 °C  
Detection: UV, 220 nm

**Peaks:**  
1. Cytosine  
2. Uracil  
3. Adenine  
4. Guanine  
5. Thymine

measurement every 14 h;  
in between flow was stopped

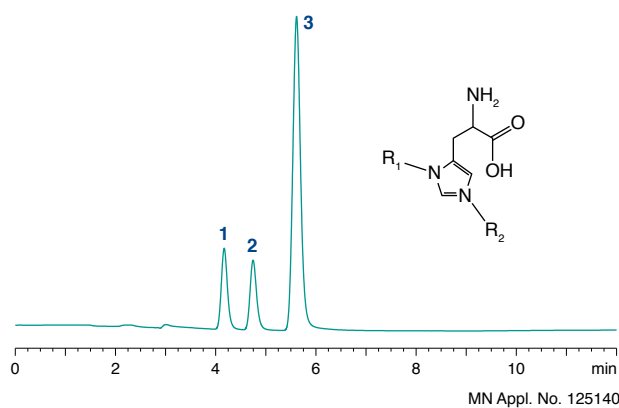


In spite of the polar character of the embedded functional group NUCLEODUR® exhibits sufficient hydrophobic properties and is very well suited for analyzing basic compounds. Good base deactivation of NUCLEODUR® PolarTec is illustrated by the separation of pyridine and phenol.

## Separation of histidine

Column: 150 x 3 mm NUCLEODUR® PolarTec, 3 µm  
Eluent: 1.0 mM perfluoropentanoic acid in water –  
0.5 mM perfluoropentanoic acid in acetonitrile  
(99.5:0.5, v/v)  
Flow rate: 0.4 mL/min  
Temperature: 20 °C  
Detection: UV, 230 nm

**Peaks:**  
1. 3-Methylhistidine R<sub>1</sub> = H, R<sub>2</sub> = CH<sub>3</sub>  
2. Histidine R<sub>1</sub> = R<sub>2</sub> = H  
3. 1-Methylhistidine R<sub>1</sub> = CH<sub>3</sub>, R<sub>2</sub> = H

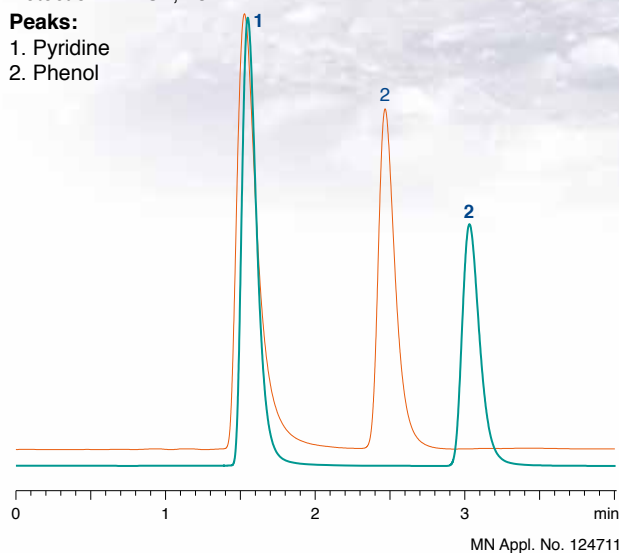


Even after days or weeks of operation in purely aqueous eluents the C<sub>18</sub> chains of NUCLEODUR® PolarTec are neither folded nor show any collapsing. A significant reduction of retention time cannot be observed.

## Phase comparison of NUCLEODUR® PolarTec

Columns: 150 x 3 mm NUCLEODUR® PolarTec, 5 µm  
150 x 3 mm Waters SymmetryShield™ RP18, 5 µm  
Eluent: acetonitrile – water (50:50, v/v)  
Flow rate: 0.56 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm

**Peaks:**  
1. Pyridine  
2. Phenol



## Key features:

- Hydrophobic phase with alternative selectivity in comparison to classical C<sub>18</sub> modifications
- Separation principle based on 4 retention mechanisms:
  - polar interactions (H bonds)
  - dipole–dipole interactions
  - π–π interactions
  - hydrophobic interactions
- Suitable for LC/MS due to low bleeding characteristics

## Technical characteristics:

Phase with pentafluorophenyl–propyl modification and multi-endcapping; pore size 110 Å; particle sizes 3 μm and 5 μm; carbon content 8%; pH stability 1–9

## Recommended application:

Aromatic and unsaturated compounds, phenols, halogenated compounds, isomers, polar compounds like pharmaceuticals, antibiotics; high retention of basic compounds

**USP L43**

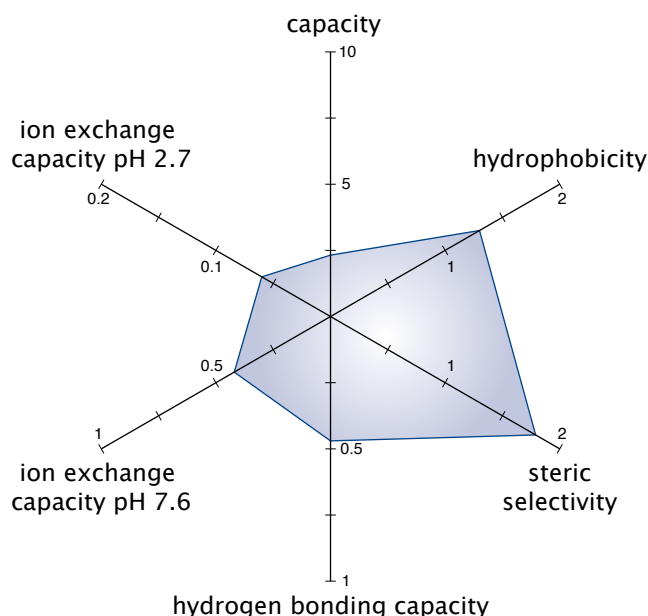
## Orthogonality in selectivity

Fluorinated stationary phases in HPLC have gained increasing interest over the last years. Most common representative of fluorinated silica phases is the pentafluorophenyl modification (PFP or F5). Especially the orthogonal selectivity compared to traditional alkyl phases widens the scope in analytical HPLC. Thus NUCLEODUR® PFP offers an excellent selectivity especially for highly polar analytes like aromatic and unsaturated compounds, phenols or halogenated hydrocarbons.

Halogen substitutes in molecules result often in an increase of their polarity accompanied by a decrease of typical retention characteristics in RP–HPLC.

While a typical C<sub>18</sub> phase just provides hydrophobic interactions between stationary phase and analyte NUCLEODUR® PFP offers four different retention mechanisms: polar interactions (H bonds), dipole–dipole interactions, π–π interactions and hydrophobic interactions. Especially the pronounced ion exchange capacity and distinct steric selectivity are typical for the character of fluorinated phases.

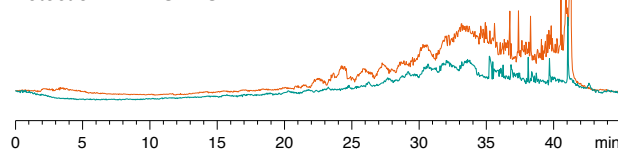
## Tanaka plot of NUCLEODUR® PFP



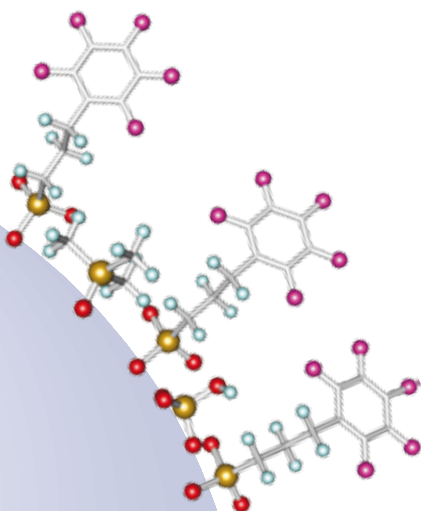
Due to low bleeding characteristics NUCLEODUR® PFP is also suitable for LC/MS.

## Bleeding of NUCLEODUR® PFP

Columns: 100 x 4.6 mm  
**NUCLEODUR® PFP, 5 μm**  
 Phenomenex Luna® PFP(2), 5 μm  
 Eluent: A) acetonitrile, B) water  
 Gradient: 5% A → 100% A in 20 min (5 min)  
 → 5% A in 5 min (5 min)  
 Flow rate: 0.2 mL/min  
 Temperature: 22 °C  
 Detection: ESI MS



MN Appl. No. 124730

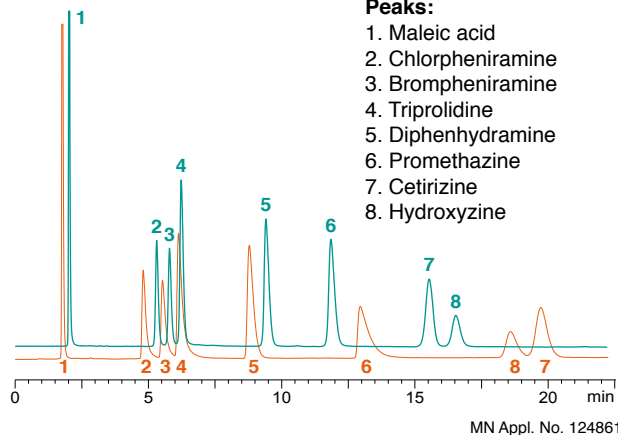


# Hydrophobic pentafluorophenyl phase

## Separation of antihistamines

Columns: 250 x 3 mm NUCLEODUR® PFP, 5 µm  
250 x 3 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub> (30:70, v/v)  
Flow rate: 0.563 mL/min  
Temperature: 30 °C  
Detection: UV, 210 nm

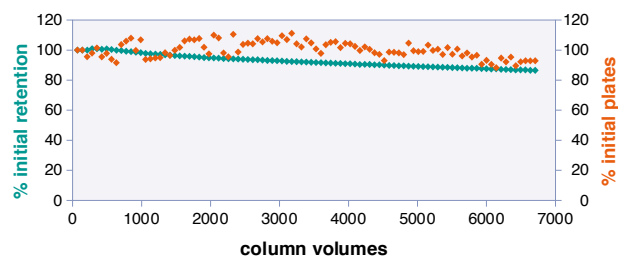
- Peaks:**
1. Maleic acid
  2. Chlorpheniramine
  3. Brompheniramine
  4. Triprolidine
  5. Diphenhydramine
  6. Promethazine
  7. Cetirizine
  8. Hydroxyzine



Based on a special surface modification procedure NUCLEODUR® PFP offers highest stability also at low pH values.

## Stability of NUCLEODUR® PFP

Column: 125 x 4 mm NUCLEODUR® PFP, 5 µm  
Eluent: acetonitrile – water, 0.1 % TFA, pH 1 (50:50, v/v)  
Flow rate: 1 mL/min  
Temperature: 80 °C  
Detection: UV, 254 nm  
Injection volume: 2 µL  
Sample: ethylbenzene

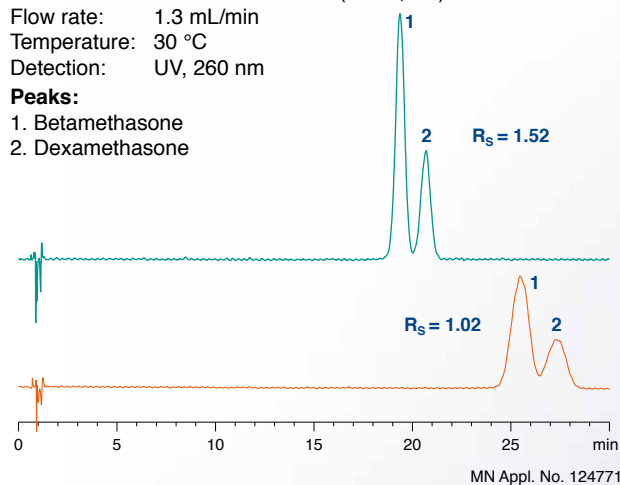


Compared to fluorinated HPLC columns of other manufacturers NUCLEODUR® PFP shows excellent separation capabilities and shorter retention times e.g., for critical isomers like beta- and dexamethasone.

## Separation of beta- and dexamethasone

Columns: 100 x 4.6 mm NUCLEODUR® PFP, 5 µm  
100 x 4.6 mm Phenomenex Luna® PFP(2), 5 µm  
Eluent: acetonitrile – water (20:80, v/v)  
Flow rate: 1.3 mL/min  
Temperature: 30 °C  
Detection: UV, 260 nm

- Peaks:**
1. Betamethasone
  2. Dexamethasone

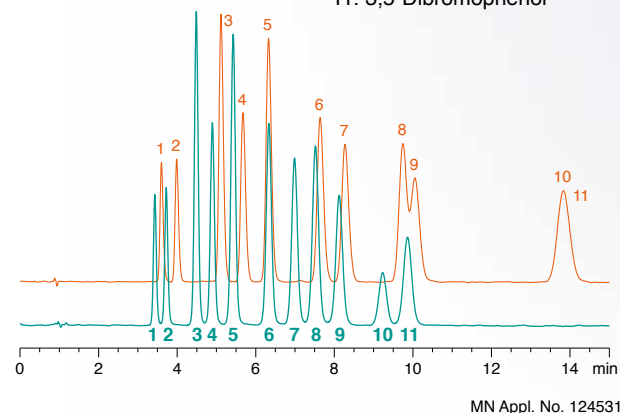


NUCLEODUR® PFP offers a completely different retention behavior compared to alkyl modified silica and is often used successfully for separations which provide just insufficient results on traditional C<sub>18</sub> phases. More and more applications in the areas of (bio-) pharma, natural compounds and environment are published and show the broad application field for fluorinated phases.

## Separation of phenol isomers

Columns: 125 x 4 mm NUCLEODUR® PFP, 5 µm  
125 x 4 mm NUCLEODUR® C<sub>18</sub> HTec, 5 µm  
Eluent: acetonitrile, 0.1 % formic acid – water, 0.1 % formic acid (35:65, v/v)  
Flow rate: 1 mL/min  
Temperature: 35 °C  
Detection: UV, 280 nm

- Peaks:**
1. *o*-Cresol
  2. *m*-Cresol
  3. 3,4-Dimethylphenol
  4. 3,5-Dimethylphenol
  5. 2,5-Dimethylphenol
  6. 2,6-Dichlorophenol
  7. 2,3-Dichlorophenol
  8. 2,4-Dichlorophenol
  9. 3,4-Dichlorophenol
  10. 2,4-Dibromophenol
  11. 3,5-Dibromophenol



# Sphinx RP

## Key features:

- Distinct selectivity based on well-balanced bifunctional surface coverage
- Widens the scope for method development based on additional  $\pi$ - $\pi$  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  $\mu\text{m}$ , 3  $\mu\text{m}$  and 5  $\mu\text{m}$ ; carbon content 15%; pH stability 1-10; high reproducibility and consistent quality

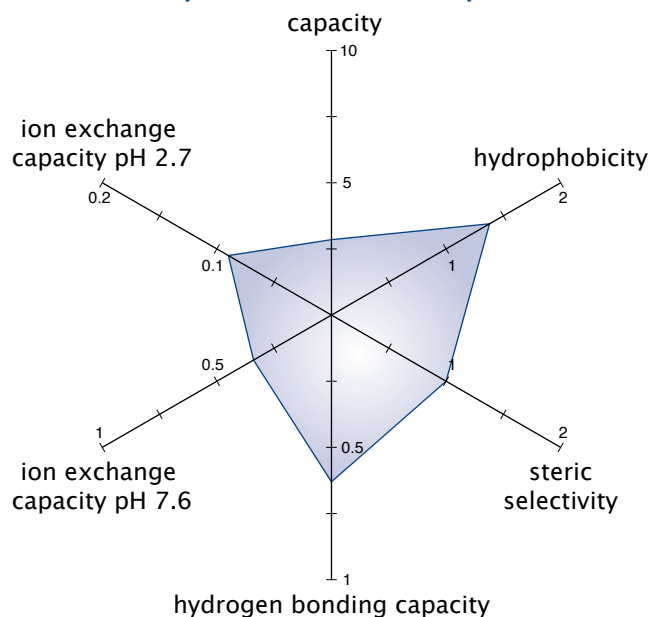
## Recommended application:

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  $\pi$ - $\pi$  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  $\text{C}_{18}$  phases. In addition, exhaustive endcapping steps minimize unwanted surface silanol activity and guarantee excellent peak shapes even for strong basic analytes.

## Tanaka plot of NUCLEODUR® Sphinx RP

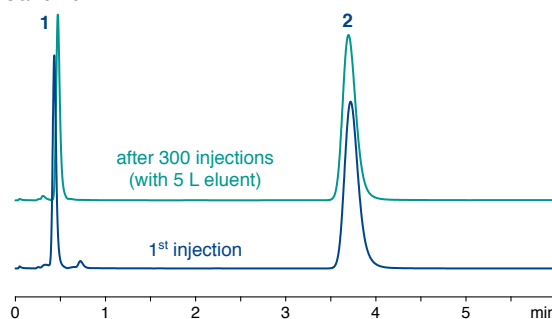


## Stability of NUCLEODUR® Sphinx RP at pH 10

Column: 50 x 4.6 mm NUCLEODUR® Sphinx RP, 5  $\mu\text{m}$   
Eluent: methanol – dil.  $\text{NH}_3$ , pH 10 (20:80, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 30 °C  
Detection: UV, 275 nm  
Injection volume: 3  $\mu\text{L}$

### 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<sub>8</sub> / C<sub>18</sub> Gravity and the polar endcapped NUCLEODUR® C<sub>18</sub> Pyramid.

The selectivity advantage of NUCLEODUR® Sphinx RP is impressively shown in the flavonoid application below.

While a baseline separation of kaempferol and isorhamnetin can be achieved on NUCLEODUR® Sphinx RP, the two compounds are not or just poorly separated on NUCLEODUR® C<sub>8</sub> Gravity or C<sub>18</sub> Gravity. The additional π-π interactions of the aromatic ring systems provide the necessary difference in retention to outperform the classical C<sub>18</sub> and C<sub>8</sub> phases.

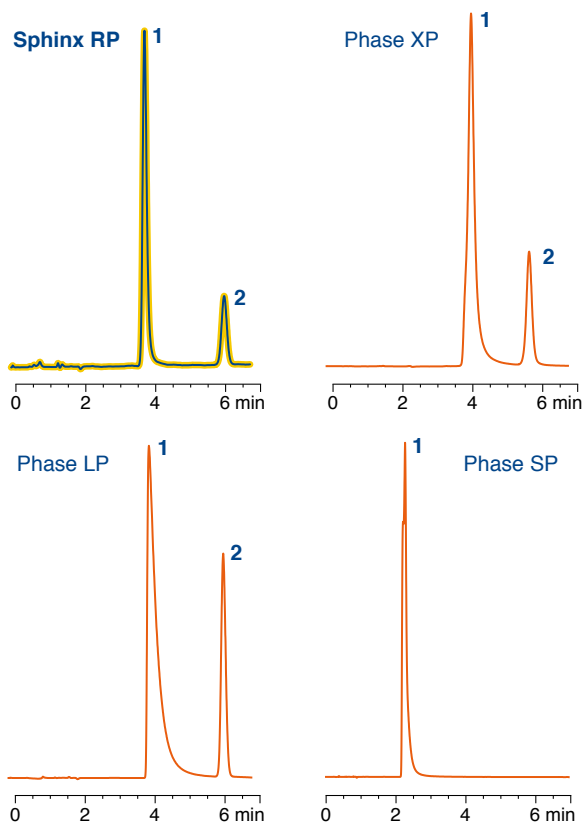
## Comparison of surface deactivation of different phenyl modified RP phases

Columns: 150 x 4.6 mm  
 NUCLEODUR® Sphinx RP, 5 μm  
 Competitor 1 (column XP)  
 Competitor 2 (column LP)  
 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

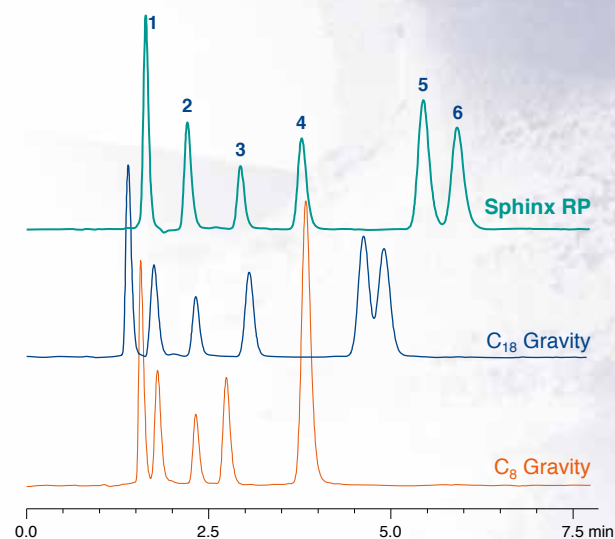
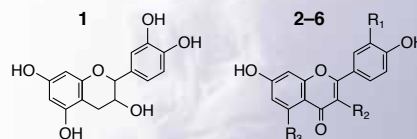
## Separation of flavonoids on 3 different NUCLEODUR® phases

Columns: 150 x 4.6 mm  
 NUCLEODUR® Sphinx RP, 5 μm  
 NUCLEODUR® C<sub>18</sub> Gravity, 5 μm  
 NUCLEODUR® C<sub>8</sub> Gravity, 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
  3. Fisetin
  4. Quercetin
  5. Kaempferol
  6. Isorhamnetin
- $R_1 = R_3 = \text{OH}, R_2 = \text{O-rutinose}$   
 $R_1 = R_2 = \text{OH}, R_3 = \text{H}$   
 $R_1 = \text{H}, R_2 = R_3 = \text{OH}$   
 $R_1 = \text{OCH}_3, R_2 = R_3 = \text{OH}$



MN Appl. No. 119830

## 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<sub>18</sub>); pore size 110 Å; particle sizes 1.8 µm, 3 µm, 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, derivatized amino acids, pesticides, fat-soluble vitamins, aldehydes, ketones and phenolic compounds  
**USP L1**

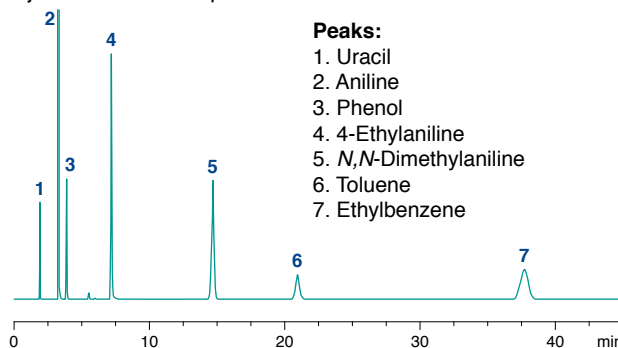
Preparative separations place high demands on silica based HPLC materials. Apart from excellent selectivity and base deactivation, robustness (pH, pressure stability, ...) and capacity are vital criteria for optimal and efficient separation at the preparative scale.

## 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<sub>18</sub> HTec scores in low bleed characteristics and is therefore highly suitable for LC/MS.

## Engelhardt test

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> HTec, 5 µm  
Eluent: methanol - water (49:51, v/v)  
Flow rate: 1 mL/min  
Temperature: 40 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

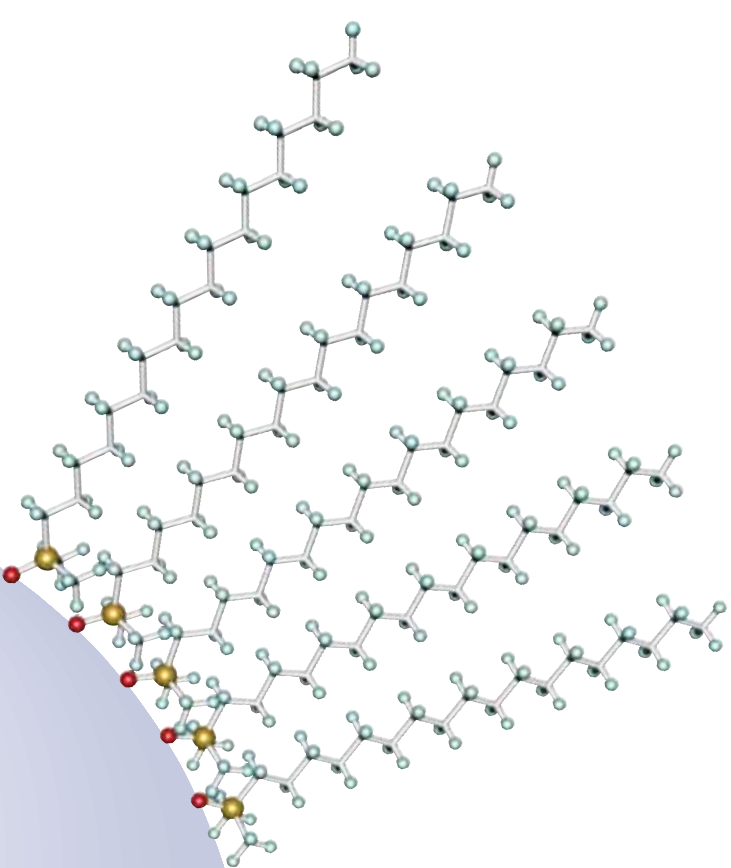
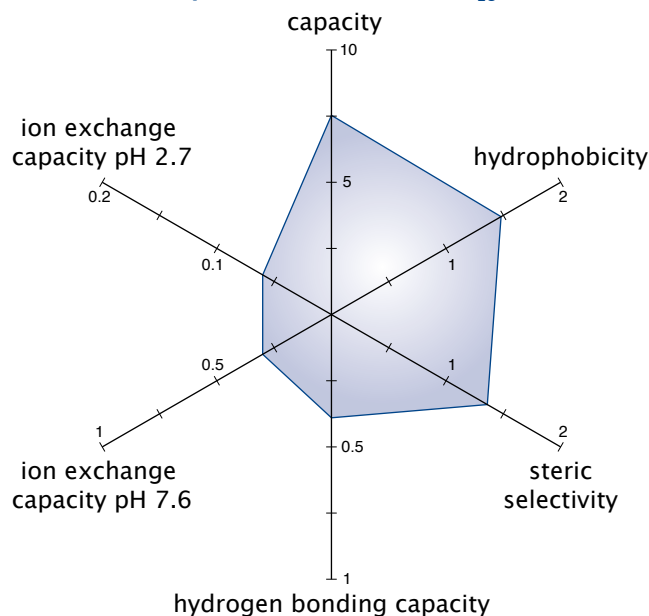


### Peaks:

- Uracil
- Aniline
- Phenol
- 4-Ethylaniline
- N,N*-Dimethylaniline
- Toluene
- Ethylbenzene

MN Appl. No. 123580

## Tanaka plot of NUCLEODUR® C<sub>18</sub> HTec





## Stability and lifetime

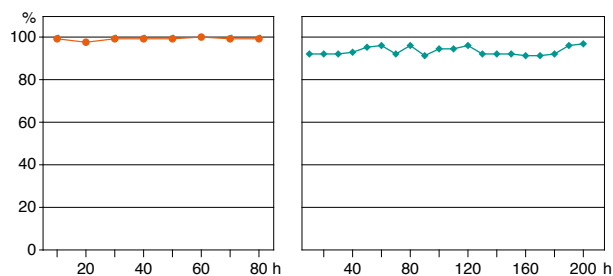
Based on fully synthetic and extremely robust totally spherical NUCLEODUR<sup>®</sup> silica, NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec offers outstanding mechanical rigidity and is thus the perfect choice also for self-packing of prep-columns. The special surface modification and endcapping procedure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

### pH stability test

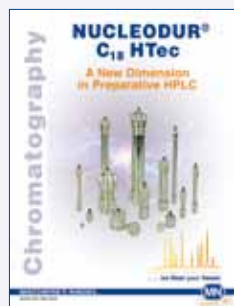
Column: 150 x 4 mm NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec, 5 μm  
Flow rate: 1 mL/min  
Detection: UV, 254 nm  
Injection: 5 μL

**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



Due to innovative surface coating procedures NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec offers excellent analytical separation properties and is the first choice for up-scaling to preparative column dimensions.



Please ask for our special NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec brochure for preparative HPLC separation.

## Capacity

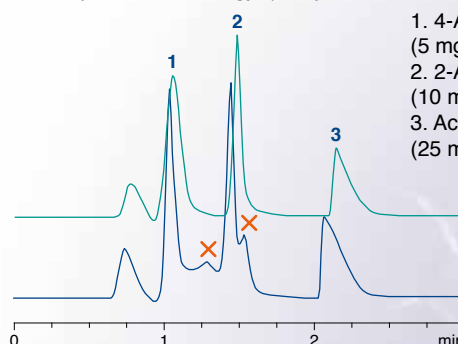
A vital criterion for efficiency in preparative HPLC is the capacity of the separation medium. NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec is characterized by a notably high loadability under both basic and acidic conditions, while competitor columns show overload effects even at lower loads (X).

### Loadability under acidic conditions

Columns: VP 100 x 21 mm NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec, 5 μm  
100 x 21.2 mm AXIA<sup>™</sup> Gemini<sup>®</sup> 5 μm C<sub>18</sub> 110 Å  
Eluent: acetonitrile – formic acid in H<sub>2</sub>O 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)



## Up-scaling

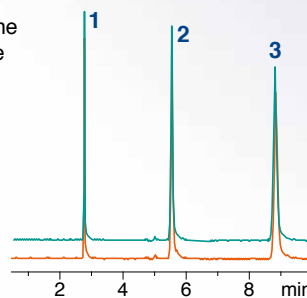
Due to highest quality standards in our silica production and phase chemistry combined with optimized packing technology, NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec delivers exceptional transferability from analytical to preparative scale. This doesn't just apply to the use of different particle sizes (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<sup>®</sup> C<sub>18</sub> HTec

Columns: EC 250 x 4.6 mm NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec, 5 μm  
VP 250 x 21 mm NUCLEODUR<sup>®</sup> C<sub>18</sub> HTec, 5 μm  
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

## Key features:

- Ideal and reliable standard RP phase for daily routine analysis and up-scaling for preparative HPLC
- Medium density octadecyl (C<sub>18</sub>) and octyl (C<sub>8</sub>) 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 C<sub>18</sub>, 10.5% for C<sub>8</sub>; pH stability 1-9, high reproducibility from lot to lot

## Recommended application:

Basic, neutral or acidic drugs  
derivatized amino acids  
pesticides  
fat-soluble vitamins  
aldehydes and ketones  
phenolic compounds  
**USP L1 (C<sub>18</sub>) / L7 (C<sub>8</sub>)**

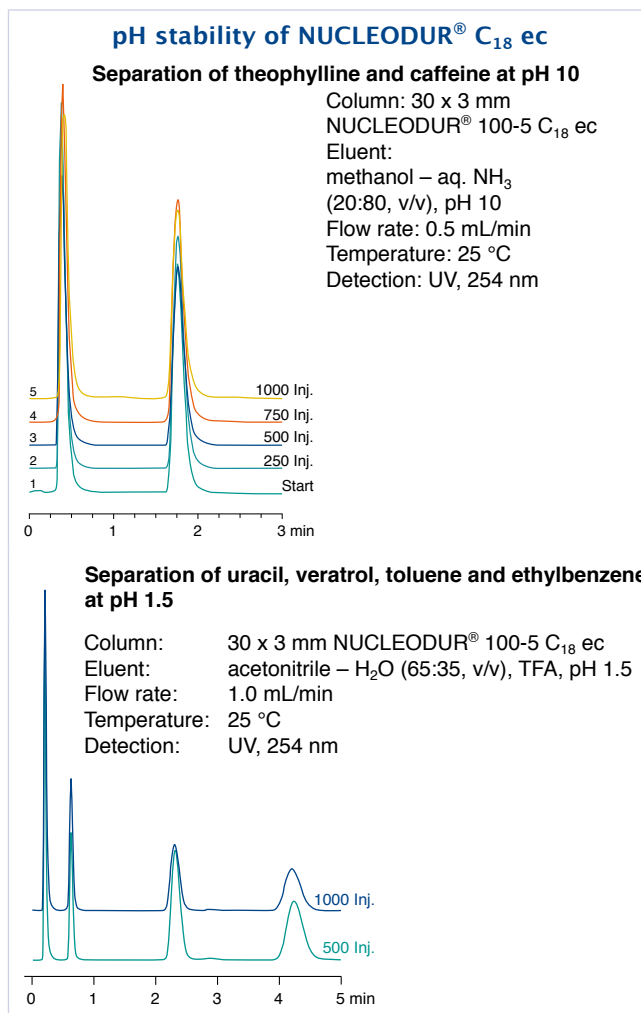
## NUCLEODUR® C<sub>18</sub> ec for daily routine analysis and up-scaling for preparative HPLC

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<sub>18</sub> 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<sub>18</sub> ec is also an ideal tool for scale-up purposes.

## 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.

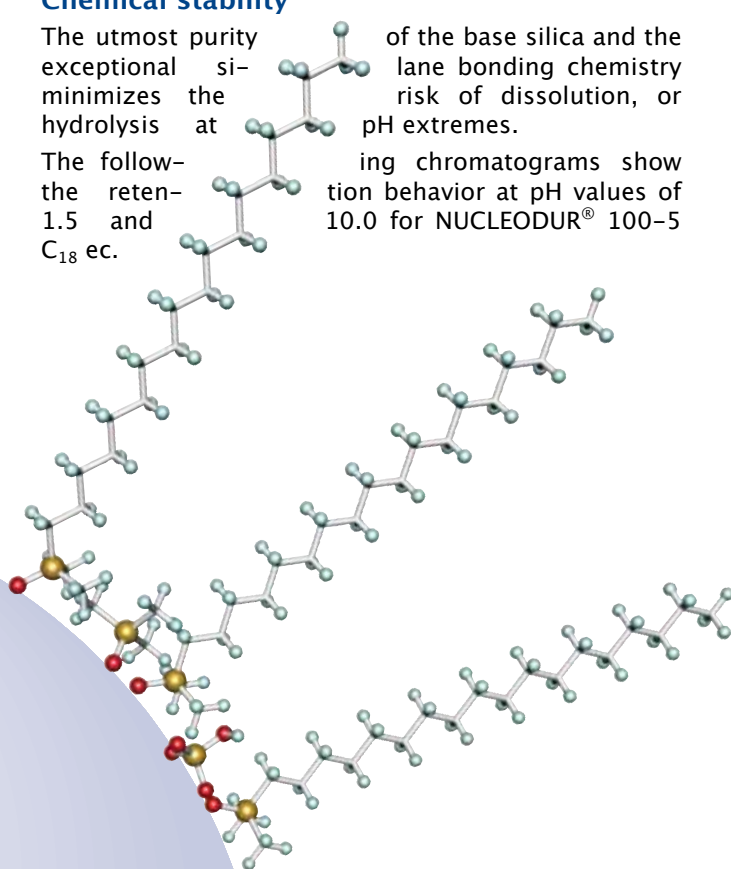
The following chromatograms show the retention behavior at pH values of 1.5 and 10.0 for NUCLEODUR® 100-5 C<sub>18</sub> ec.



## Loadability

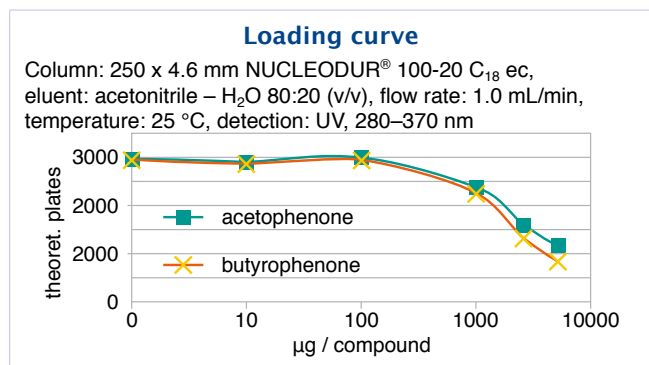
Loadability, probably the most important feature for preparative LC applications, is determined by pore size, pore volume and surface area of the packing. However, it can also be influenced by the molecular weight of the analytes. In the figure below the mass loading curve for acetophenone and butyrophenone on a NUCLEODUR®

NUCLEODUR®

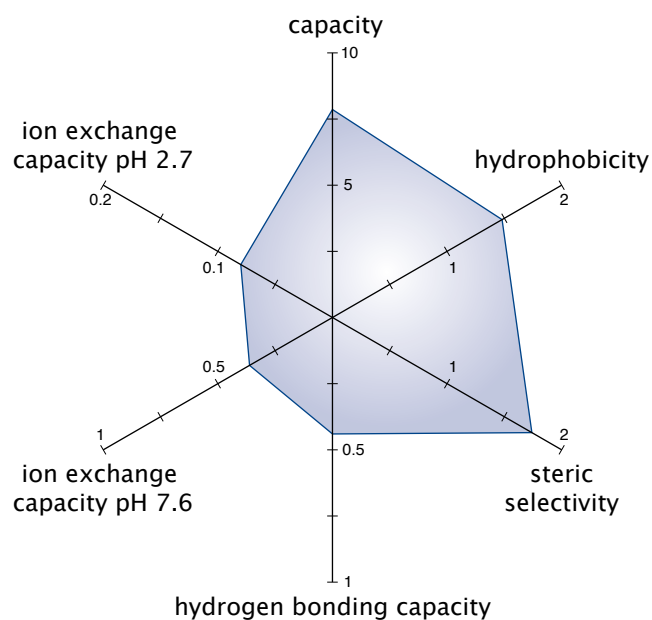


# Nonpolar phases for routine analyses

100–20 C<sub>18</sub> ec column describes the correlation between the increase of column loading and the decrease of separation efficiency.



## Tanaka plot of NUCLEODUR® C<sub>18</sub> ec



## NUCLEODUR® octyl phases

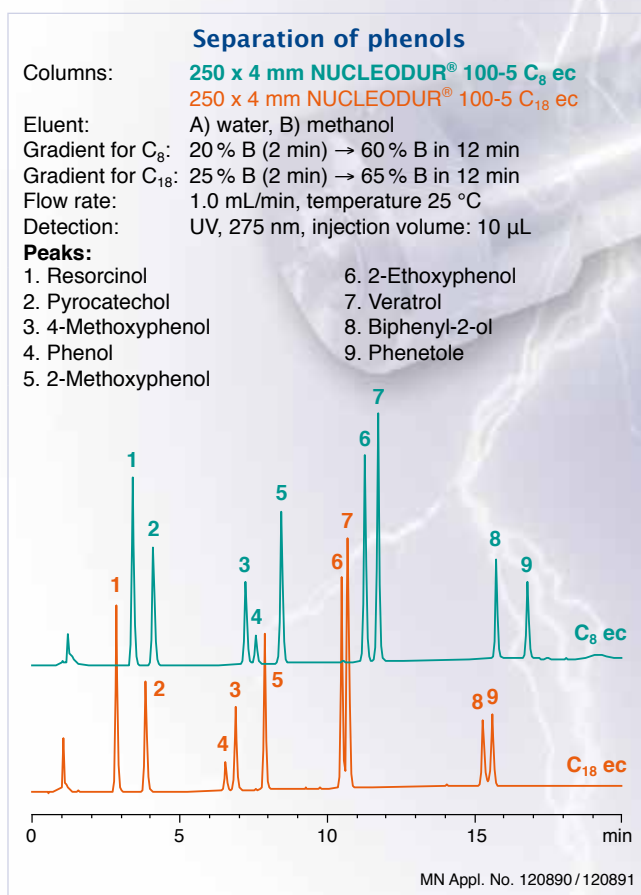
In addition to the program of NUCLEODUR® C<sub>18</sub> phases MACHEREY-NAGEL offers the corresponding octyl modified NUCLEODUR® C<sub>8</sub> Gravity and NUCLEODUR® C<sub>8</sub> ec columns to expand the reversed phase tool box effectively. Based on the same totally spherical and highly pure silica the C<sub>8</sub> phases exhibit the same excellent chemical and mechanical stability features as the C<sub>18</sub> counterparts. Indeed NUCLEODUR® C<sub>8</sub> 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<sub>18</sub> phases).

NUCLEODUR® C<sub>8</sub> ec and NUCLEODUR® C<sub>8</sub> Gravity are most suitable for the development of new methods but also for robust routine analysis.

## C<sub>18</sub> or C<sub>8</sub> · the best of both worlds

Chromatographers now might wonder about the differences between C<sub>8</sub> and C<sub>18</sub> 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<sub>8</sub> ec and NUCLEODUR® C<sub>18</sub> ec. The separation of phenols below 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.



## Some general principles are:

- High density C<sub>8</sub> and C<sub>18</sub> phases allow tailing-free elution, also for very polar compounds
- Octyl phases (C<sub>8</sub>) show superior polar selectivity
- Octadecyl phases (C<sub>18</sub>) show superior hydrophobic selectivity
- Hydrophobic compounds show shorter retention times on C<sub>8</sub> phases

## 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

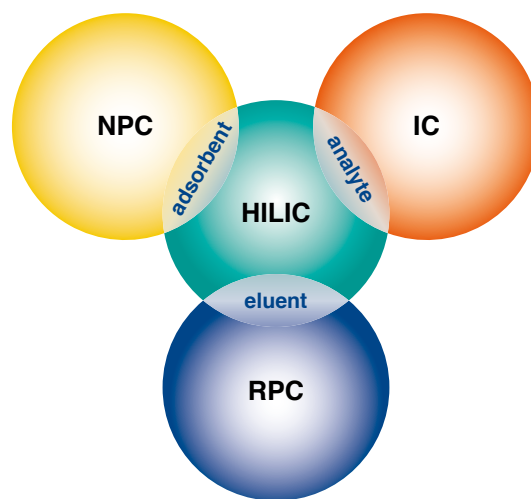
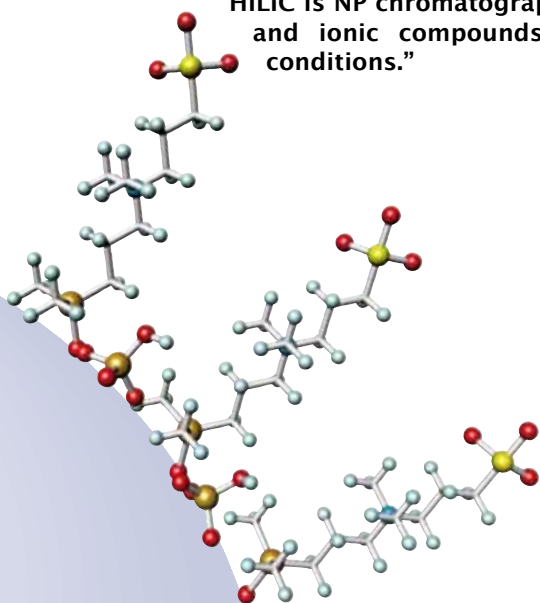
Separation science is always looking for new and effective strategies to accomplish the tasks of modern analytics. Especially for polar compounds reversed phase HPLC – the most common analytical method – is often limited. Here, hydrophilic stationary phases provide an additional tool for the separation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

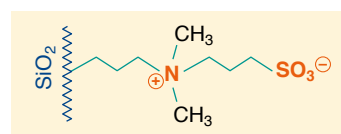
HILIC combines the characteristics of the 3 major methods in liquid chromatography – reversed phase (RPC), normal phase (NPC) and ion chromatography (IC):

- stationary phases (adsorbents) are mostly polar modifications of silica or polymers (SiOH, NH<sub>2</sub>, Diol, (zwitter) ions, ...) – like in NPC
- mobile phases (eluent) 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 equalization and in an overall neutrally charged but highly polar surface.

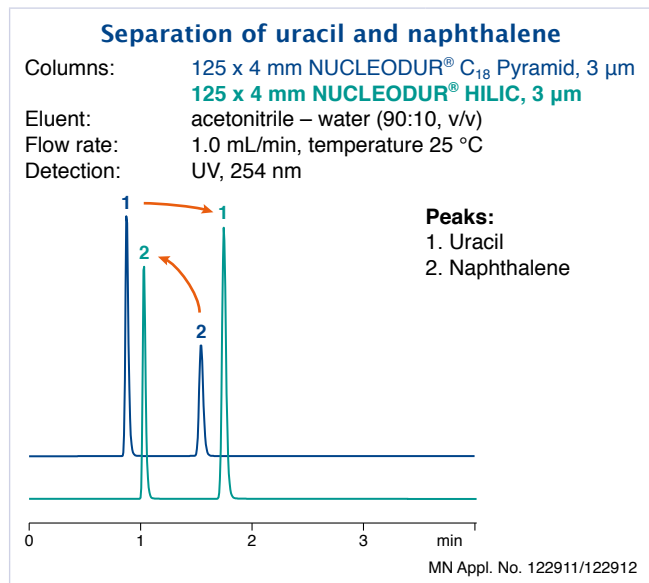


## Retention characteristic

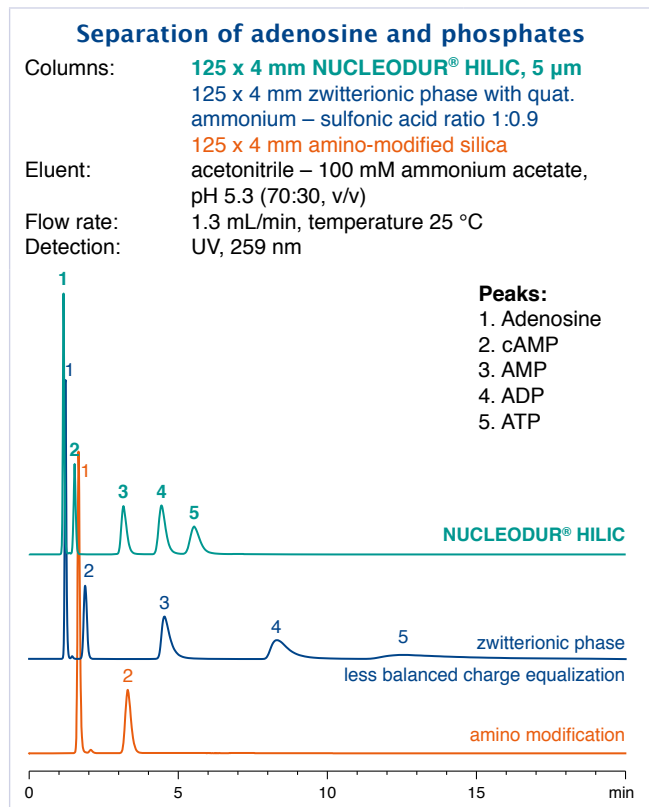
Commonly HILIC is described as partition chromatography or liquid/liquid extraction system between the mobile and stationary phase. Versus a water-poor mobile phase a water-rich layer on the surface of the polar stationary phase is formed. Thus, a distribution of the analytes between these two layers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

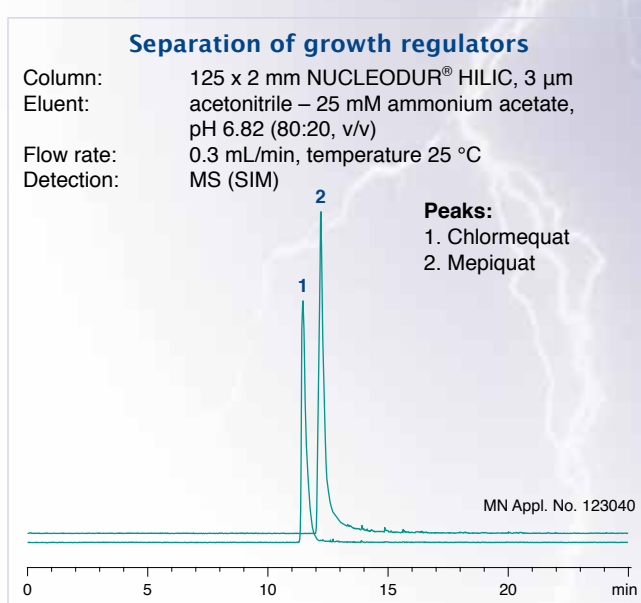
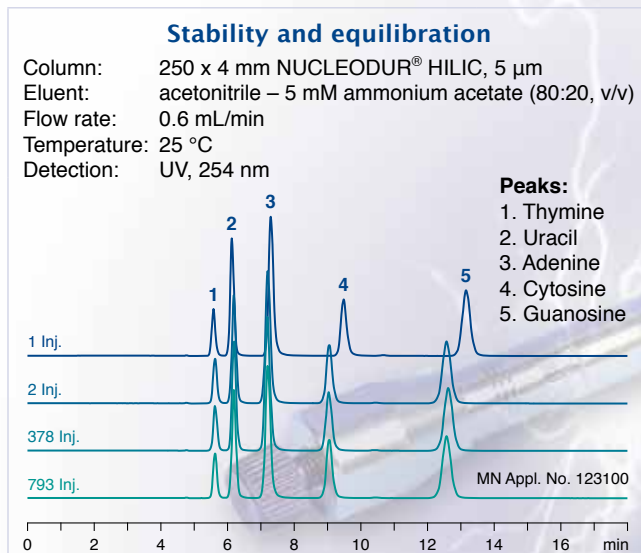


In comparison with medium polar aminopropyl phases or modification with less balanced charge equalization NUCLEODUR® HILIC shows a superb separation and peak shape for critical compounds like adenosine phosphates.



## 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 2<sup>nd</sup> 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.

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

## 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; carbon content 7%; special endcapping; high reproducibility from lot to lot; different retention characteristics in comparison to C<sub>8</sub> and C<sub>18</sub>

## 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<sub>18</sub> or C<sub>8</sub> 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 endcapped and highly reproducible (see figure top right) NUCLEODUR® CN-RP phase has cyanopropyl groups on the surface able to generate a clearly recognizable different retention behavior compared to purely alkyl-functionalized surface modifications (see figure down right).

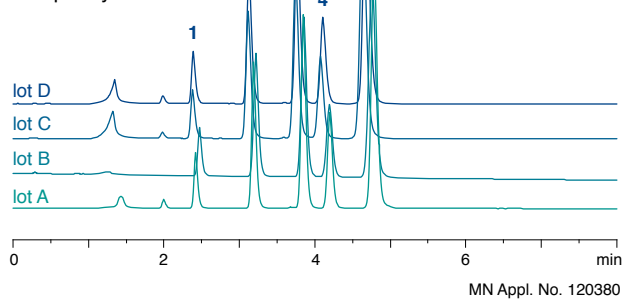
The polarity of the NUCLEODUR® CN-RP phase can be classified as intermediate based on multiple retention mechanisms such as dipole-dipole, π-π, and also hydrophobic interactions [C. S. Young and R. J. Weigand, LCGC 20 (2002) 464–473]. Therefore, this phase shows a distinct selectivity for polar organic 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)].

## Reproducibility of NUCLEODUR® CN-RP

Column: 250 x 4 mm NUCLEODUR® 100-5 CN-RP  
Eluent: acetonitrile – water (60:40, v/v)  
Flow rate: 1.0 mL/min, temperature 20 °C  
Detection: UV, 254 nm, injection volume: 5 µL

### Peaks:

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

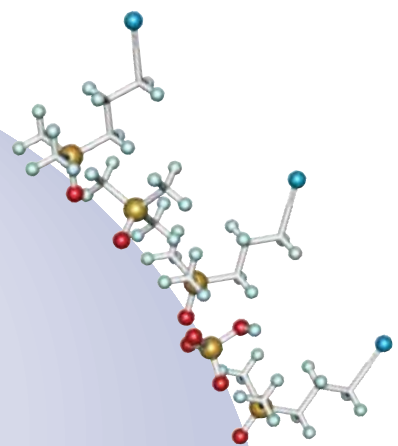
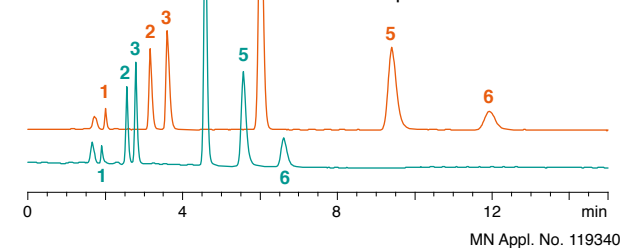


## Separation of cold medicine ingredients on two different NUCLEODUR® phases

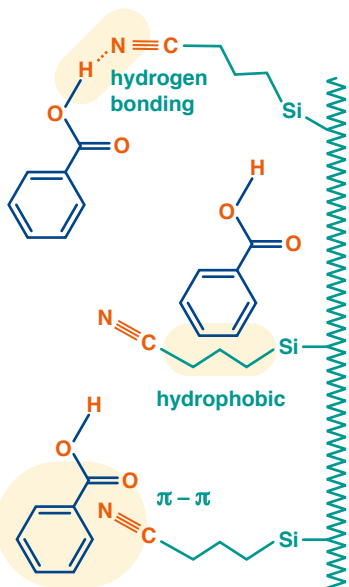
Columns: 250 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
250 x 4 mm NUCLEODUR® 100-5 CN-RP  
Eluent: acetonitrile – 100 mM sodium citrate, pH 2.5 (15:85, v/v)  
Flow rate: 1.0 mL/min, temperature 25 °C  
Detection: UV, 270 nm, injection volume: 10 µL

### Peaks:

1. Maleic acid
2. Norephedrine
3. Ephedrine
4. Paracetamol
5. Chlorpheniramine
6. Brompheniramine



## Interactions on cyano-modified silica



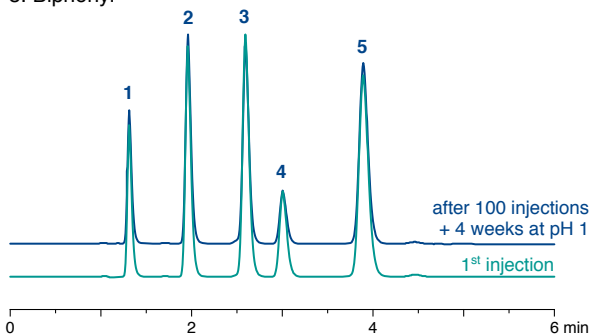
Short-chain bonded phases are sometimes suspected of revealing shortcomings in stability towards hydrolysis at low pH [J. J. Kirkland, LCGC 14 (1996) 486–500]. The following chromatograms show that even after 100 sample injections and four weeks storage at pH 1 (blue curve), neither a considerable shift in retention, nor a visible change in peak symmetry could be noticed (green curve = new column).

## 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  $\mu$ L

### 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.

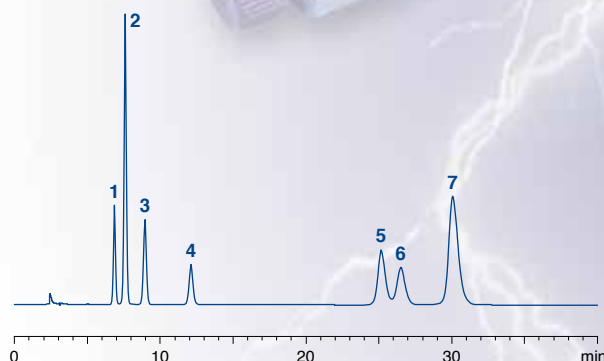
## Separation of steroids in normal phase and reversed phase mode

### 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  $\mu$ L

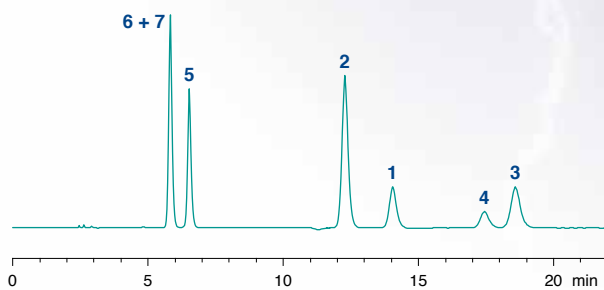
### Peaks:

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

### 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; carbon content 2.5%; 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

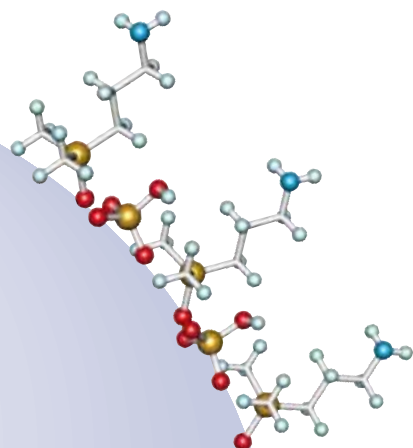
Some compounds, especially polar substances, cannot be sufficiently resolved on C<sub>18</sub> phases. Polar-modified silica phases offer alternative selectivities such as expanding the spectrum of analytical HPLC into the polar range.

### 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® NH<sub>2</sub>, 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® NH<sub>2</sub> is the separation of simple and complex sugars, sugar alcohols and other hydroxy compounds under RP conditions as well as hydrocarbons under NP conditions.

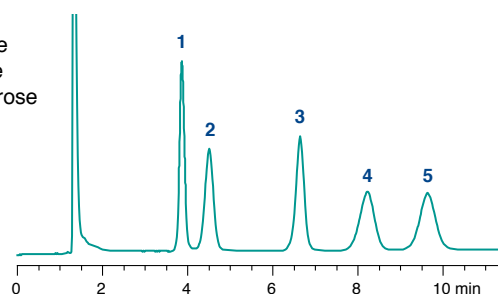


### Reversed phase separation of sugars

Column: 250 x 4 mm NUCLEODUR® 100-5 NH<sub>2</sub>-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



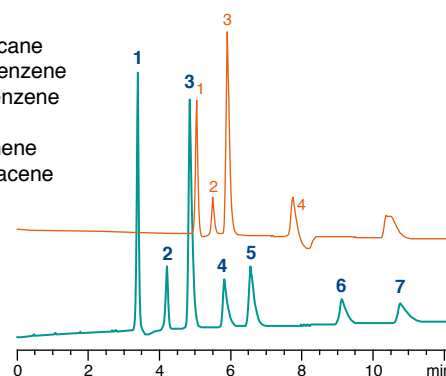
MN Appl. No. 122160

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

Columns: 250 x 4 mm NUCLEODUR® 100-5 NH<sub>2</sub>  
conventional aminopropyl phase  
Eluent: heptane  
Flow rate: 1 mL/min  
Detection: RI

#### Peaks:

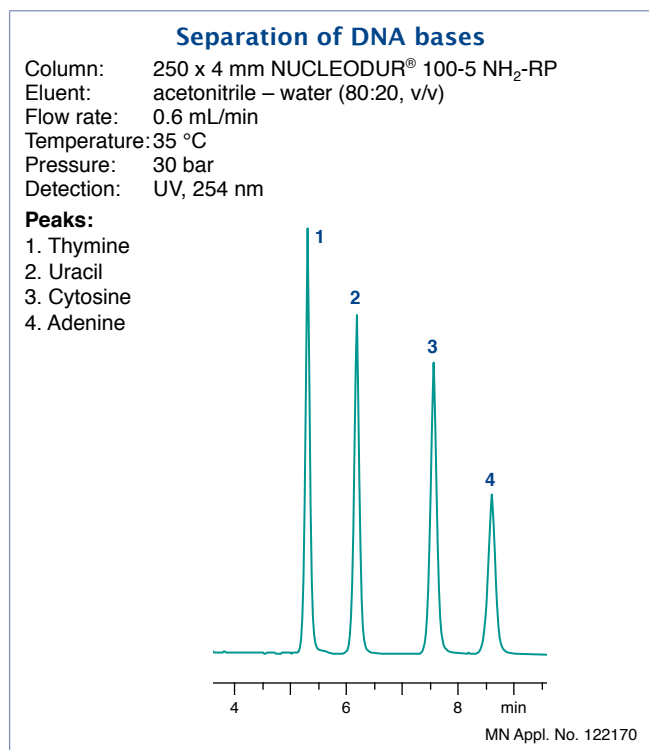
1. Cyclohexane
2. 1-Phenyldodecane
3. 1,2-Dimethylbenzene
4. Hexamethylbenzene
5. Naphthalene
6. Dibenzothiophene
7. 9-Methylanthracene



MN Appl. No. 122180

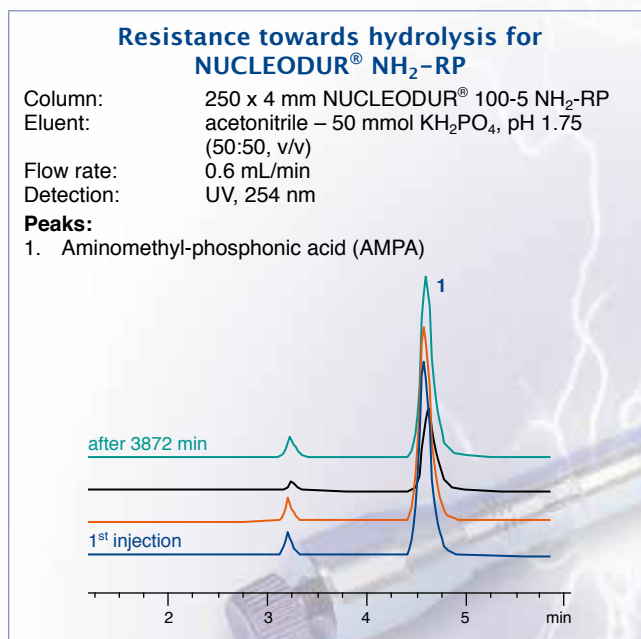


Even at lower flow rates than for C<sub>18</sub> phases, NUCLEODUR® NH<sub>2</sub> achieves good separations of polar compounds such as DNA bases – this reduces the back pressure as well as the solvent consumption. Even very polar compounds like streptomycin are retained sufficiently for quantitative and qualitative analysis.



One of the main problems with conventional amino phases is insufficient resistance towards hydrolysis. Due to a special method of surface modification NUCLEODUR® NH<sub>2</sub> 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.

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, glufonate, ...) – you may find the complete application 122190 in the "Applications" section on page 60.



Based on the superspherical silica NUCLEODUR® this amino 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<sub>2</sub> offers the advantage of reliable analyses especially for routine work.

## SiOH

### Key features:

- Totally spherical high purity silica
- Pressure stable up to 600 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<sup>2</sup>/g; pH stability 2–8; metal content < 10 ppm (see table on page 1)

### Recommended application:

Polar and mid-polar compounds under normal phase conditions  
 USP L3

# Applications

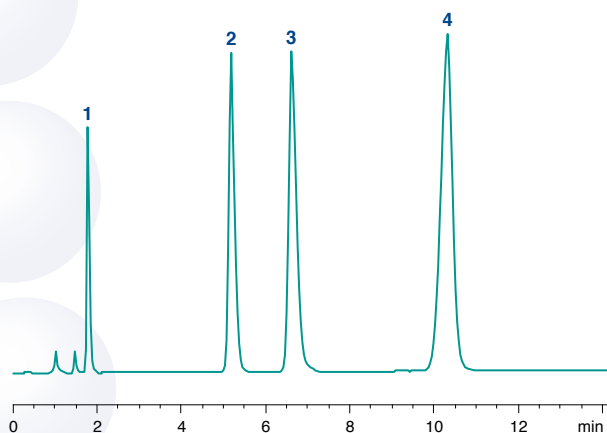
## Anesthetics

### MN Appl. No. 119410

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
Eluent: methanol – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 6.95 (65:35, v/v)  
Flow rate: 1 mL/min  
Temperature: 30 °C  
Detection: UV, 254 nm  
Injection volume: 13 µL

#### Peaks:

1. Benzocaine
2. Lidocaine
3. Tetracaine
4. Butacaine



## Analgesics

### MN Appl. No. 118600

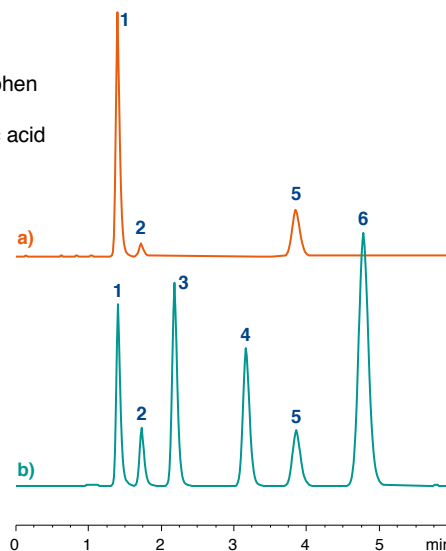
Column: 125 x 4 mm NUCLEODUR® C<sub>8</sub> Gravity, 5 µm  
Eluent: methanol – 0.1 % phosphoric acid (40:60, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 240 nm

a) Thomapyrin® tablet; b) standard

Thomapyrin® is a trademark of Boehringer Ingelheim Pharma KG

#### Peaks:

1. Paracetamol
2. Caffeine
3. 2-Acetamidophen
4. Acetanilide
5. Acetylsalicylic acid
6. Phenactin



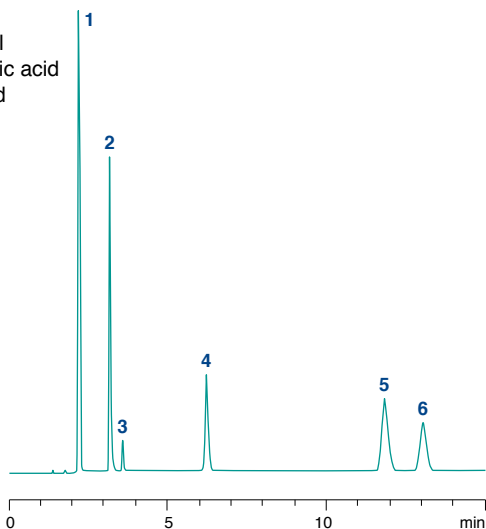
## Analgesics

### MN Appl. No. 117770

Column: 250 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 2.5 (50:50, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 230 nm  
Injection volume: 5 µL

#### Peaks:

1. Paracetamol
2. Acetylsalicylic acid
3. Salicylic acid
4. Ketoprofen
5. Diclofenac
6. Ibuprofen



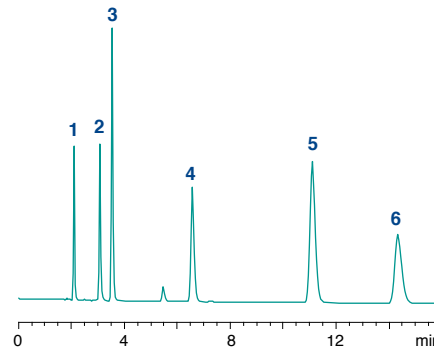
## Analgesics

### MN Appl. No. 119160

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
Eluent: acetonitrile – 0.1 % TFA (50:50, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

#### Peaks:

1. Paracetamol
2. Acetylsalicylic acid
3. Methyl-4-hydroxybenzoate
4. Ketoprofen
5. Flurbiprofen
6. Ibuprofen



For a fast separation (< 1 min) of acetylsalicylic acid and salicylic acid on NUCLEODUR® 100-5 C<sub>18</sub> ec see application 117780 at [www.mn-net.com](http://www.mn-net.com).

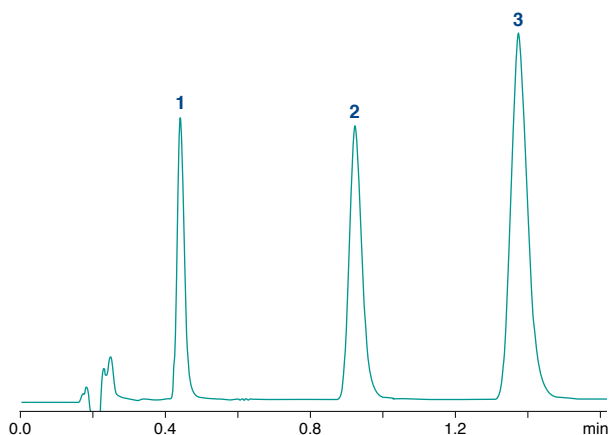
## Anti-inflammatory drugs

**MN Appl. No. 122130**

Column: 50 x 3 mm NUCLEODUR® C<sub>18</sub> Pyramid, 1.8 µm  
 Eluent: phosphate buffer, pH 2.5 – acetonitrile – methanol (425:475:100, v/v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 50 °C  
 Detection: UV, 240 nm  
 Injection volume: 2 µL

**Peaks:**

1. Chlorocresol
2. Clobetasol 17-propionate
3. Beclometasone dipropionate



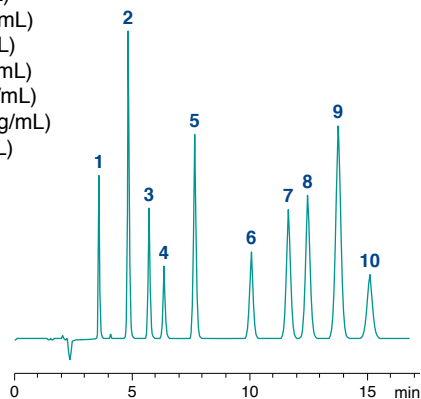
## Analgesic and anti-inflammatory drugs

**MN Appl. No. 118590**

Column: 250 x 4 mm NUCLEODUR® 100-5 C<sub>8</sub> ec  
 Eluent: acetonitrile – 1 % acetic acid (48:52, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 230 nm  
 Injection volume: 10 µL

**Peaks:**

1. Acetylsalicylic acid (1.6 µg/mL)
2. Tolmetin (26 µg/mL)
3. Piroxicam (26 µg/mL)
4. Suprofen (26 µg/mL)
5. Naproxen (0.64 µg/mL)
6. Diflunisal (1.6 µg/mL)
7. Fenoprofen (26 µg/mL)
8. Flurbiprofen (26 µg/mL)
9. Indomethacin (52 µg/mL)
10. Ibuprofen (52 µg/mL)



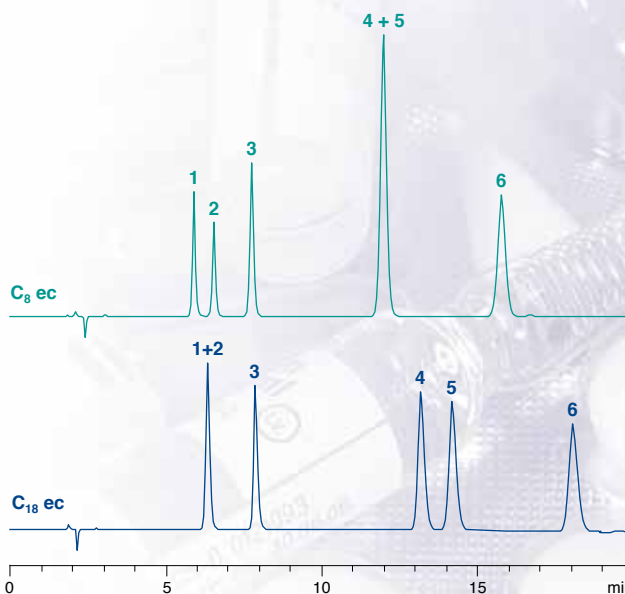
## Anti-inflammatory drugs

**MN Appl. No. 120880/120881**

Columns: 50 x 4 mm NUCLEODUR® 100-5 C<sub>8</sub> ec  
 250 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
 Eluent: acetonitrile – water, 1 % acetic acid (48:52, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 230 nm  
 Injection volume: 10 µL

**Peaks:**

1. Piroxicam
2. Suprofen
3. Ketoprofen
4. Carprofen
5. Fenoprofen
6. Diclofenac



This separation of various non-steroidal anti-inflammatory drugs illustrates the differences in polarity between C<sub>8</sub> and C<sub>18</sub> and the resulting impact on efficiency. NUCLEODUR® C<sub>8</sub> ec exhibits enhanced selectivity and excellent resolution for the polar compounds piroxicam and suprofen which co-elute on the C<sub>18</sub> column. However due to the longer alkyl chain NUCLEODUR® C<sub>18</sub> ec shows a distinct hydrophobic selectivity that leads to baseline separation of the more non-polar analytes carprofen and fenoprofen with superior peak shapes.

# Applications

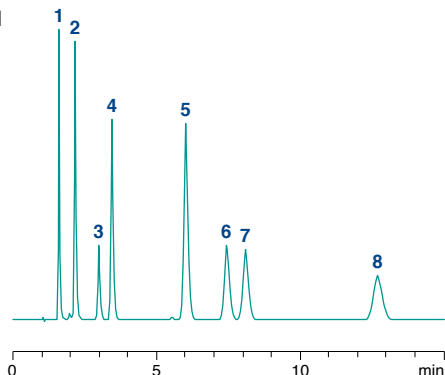
## Anti-inflammatory drugs

MN Appl. No. 117830

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 2.5 (45:55, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 22 °C  
Detection: UV, 230 nm  
Injection volume: 5 µL

### Peaks:

1. Acetylsalicylic acid
2. Sulindac
3. Tolmetin
4. Ketoprofen
5. Flurbiprofen
6. Diclofenac
7. Ibuprofen
8. Meclofenamic acid

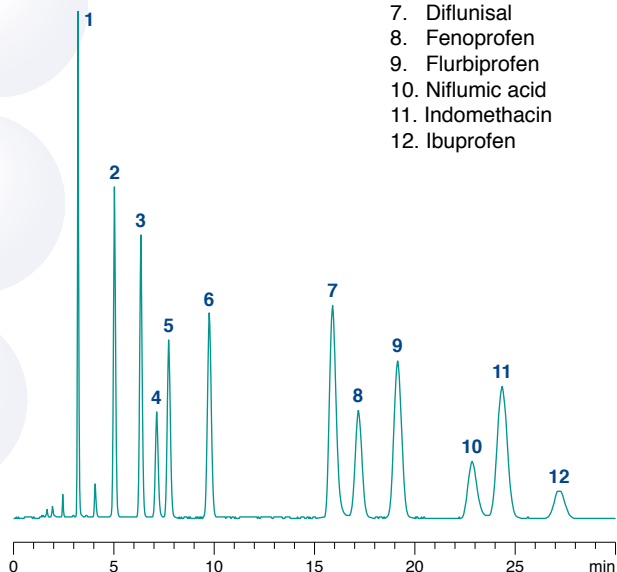


MN App. No. 122550

Column: 250 x 4.6 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Flow rate: 1.3 mL/min  
other conditions as above

### Peaks:

1. Acetylsalicylic acid
2. Sulindac
3. Piroxicam
4. Suprofen
5. Tolmetin
6. Naproxen
7. Diflunisal
8. Fenoprofen
9. Flurbiprofen
10. Niflumic acid
11. Indomethacin
12. Ibuprofen



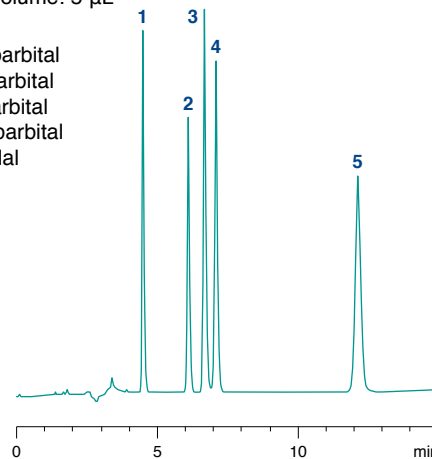
## Barbiturates

MN Appl. No. 117820

Column: 250 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
Eluent: acetonitrile – water (50:50, v/v)  
Flow rate: 0.7 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

### Peaks:

1. Phenobarbital
2. Pentobarbital
3. Hexobarbital
4. Mephobarbital
5. Thiamylal



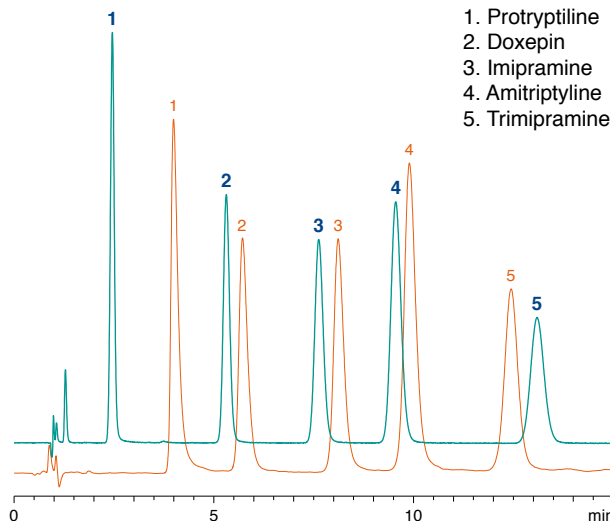
## Tricyclic antidepressants

MN Appl. No. 124622

Columns: 150 x 3 mm NUCLEODUR® PolarTec, 5 µm  
150 x 3 mm Waters SymmetryShield™ RP18, 5 µm  
Eluent: methanol – 25 mM KH<sub>2</sub>PO<sub>4</sub>, pH 7 (70:30, v/v)  
Flow rate: 0.66 mL/min  
Temperature: 30 °C  
Detection: UV, 254 nm  
Injection volume: 1 µL

### Peaks:

1. Protryptiline
2. Doxepin
3. Imipramine
4. Amitriptyline
5. Trimipramine



Excellent endcapping of NUCLEODUR® PolarTec displays significantly better peak shapes and less tailing for strong basic components compared to other phases with embedded polar group.

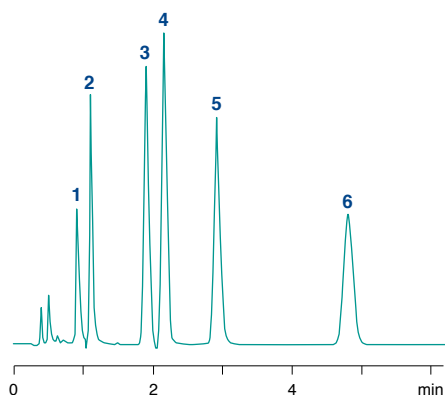
## Tricyclic antidepressants

### MN Appl. No. 117800

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
 Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 7.0 (65:35, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm  
 Injection volume: 2 µL

#### Peaks:

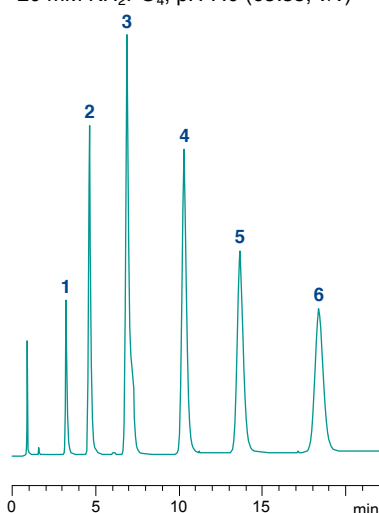
1. Protriptyline
2. Nortriptyline
3. Doxepin
4. Imipramine
5. Amitriptyline
6. Trimipramine



### MN Appl. No. 118520

Column: 125 x 4 mm NUCLEODUR® C<sub>8</sub> Gravity, 5 µm  
 Eluent: methanol – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 7.0 (65:35, v/v)  
 Temperature: 25 °C

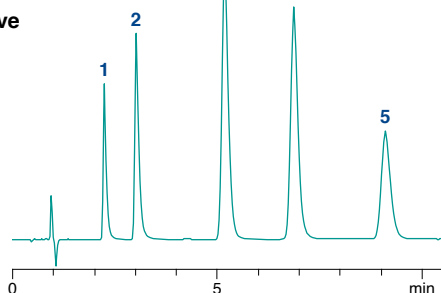
#### Peaks and other conditions as above



### MN Appl. No. 119200

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
 Eluent: methanol – 20 mM NH<sub>4</sub>H<sub>2</sub>PO<sub>4</sub>, pH 6.95 (70:30, v/v)  
 Temperature: 40 °C  
 Injection volume: 5 µL

#### Peaks and other conditions as above



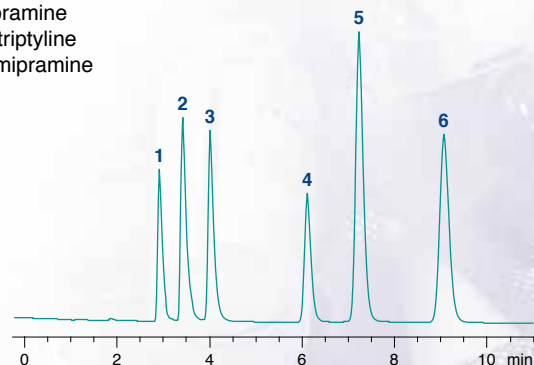
## Tricyclic antidepressants

### MN Appl. No. 121210

Column: 150 x 4 mm NUCLEODUR® C<sub>18</sub> Isis, 5 µm  
 Eluent: methanol – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 7 (75:25, v/v)  
 Flow rate: 1 mL/min  
 Temperature: 40 °C  
 Detection: UV, 230 nm  
 Injection volume: 8 µL

#### Peaks:

1. Protriptyline
2. Maprotiline
3. Nortriptyline
4. Imipramine
5. Amitriptyline
6. Clomipramine



Peak symmetry at 10 % of peak height:

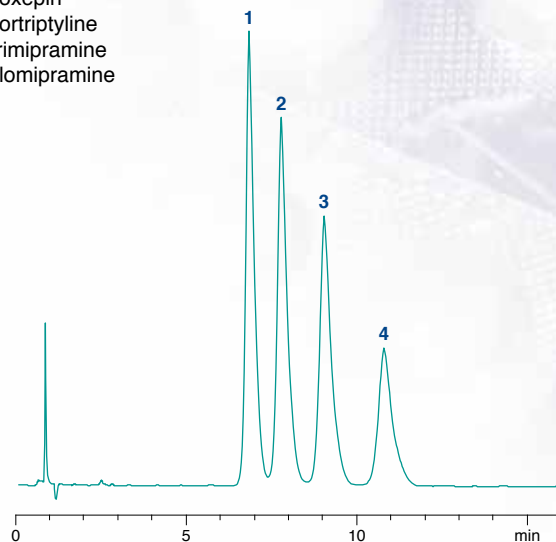
A <sub>s</sub> (imipramine):	1.29
A <sub>s</sub> (amitriptyline):	1.26
A <sub>s</sub> (clomipramine):	1.16

### MN Appl. No. 119280

Column: 250 x 4 mm NUCLEODUR® 100-5 CN-RP  
 Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 6.5 (55:45, v/v)  
 Flow rate: 1 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm  
 Injection volume: 2.5 µL (25 µg/mL)

#### Peaks:

1. Doxepin
2. Nortriptyline
3. Trimipramine
4. Clomipramine



# Applications

## Neuroleptics

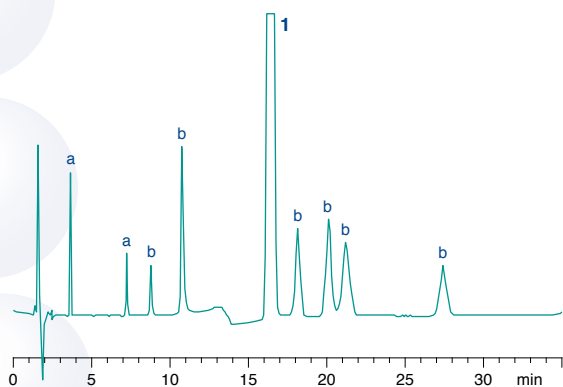
### MN Appl. No. 121612

Column: 250 x 4 mm NUCLEODUR® C<sub>8</sub> Gravity, 5 µm  
Eluent: acetonitrile – 6.0 g/L KH<sub>2</sub>PO<sub>4</sub>, 2.9 g/L sodium dodecylsulfate, 9.0 g/L tetra-*n*-butylammonium bromide pH 8 (40:60, v/v)

Flow rate: 1.5 mL/min  
Temperature: 40 °C  
Detection: 237 nm  
Injection volume: 5 µL

#### Peaks:

1. Chlorprothixene hydrochloride
- a. Additives
- b. Impurities



For separation on NUCLEODUR® C<sub>18</sub> Gravity see application 121611 at [www.mn-net.com](http://www.mn-net.com).

## Cold medicine

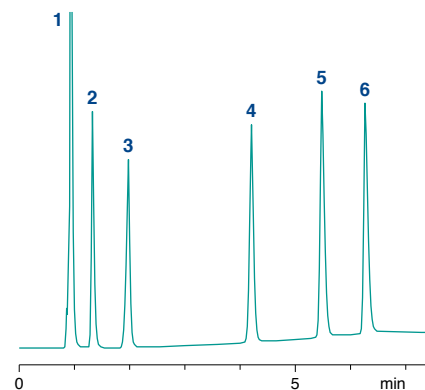
### MN Appl. No. 117810

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluents: A) 50 mM KH<sub>2</sub>PO<sub>4</sub> + 5 mM pentanesulfonate (Na salt), pH 2.5; B) methanol  
35% B → 55% B in 5 min

Flow rate: 1.0 mL/min  
Temperature: 40 °C  
Detection: UV, 230 nm  
Injection volume: 5 µL

#### Peaks:

1. Maleic acid
2. Paracetamol
3. Pseudoephedrine
4. Benzoic acid
5. Chlorpheniramine
6. Dextromethorphan



## Gastric acid inhibitors

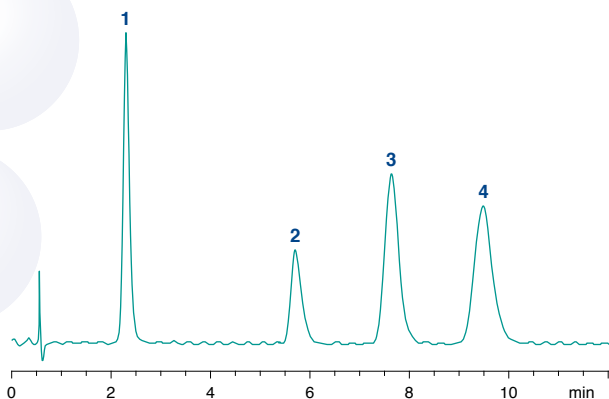
### MN Appl. No. 122520

Column: 75 x 4.6 mm NUCLEODUR® C<sub>18</sub> Gravity, 3 µm  
Eluent: methanol – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 7 with TEA (20:80, v/v)

Flow rate: 1.3 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 10 µL

#### Peaks:

1. Famotidine
2. Cimetidine
3. Nizatidine
4. Pirenzepine hydrochloride



## Cold medicine ingredients

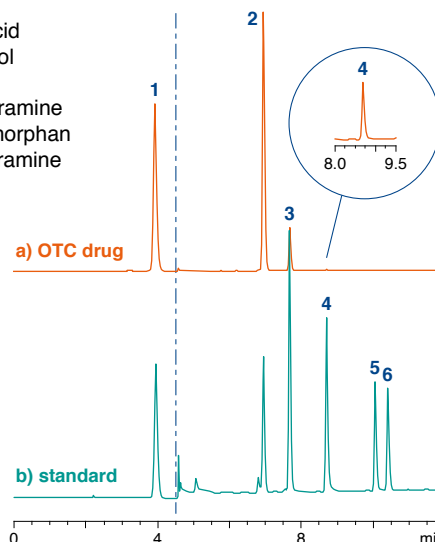
### MN Appl. No. 119110/119120

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
Eluent: A) 50 mM NH<sub>4</sub>H<sub>2</sub>PO<sub>4</sub>, pH 2.5; B) acetonitrile  
0% B → 60% B in 13 min

Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 230 nm for 4.5 min, then 261 nm  
Injection volume: a) 2 µL, b) 4 µL

#### Peaks:

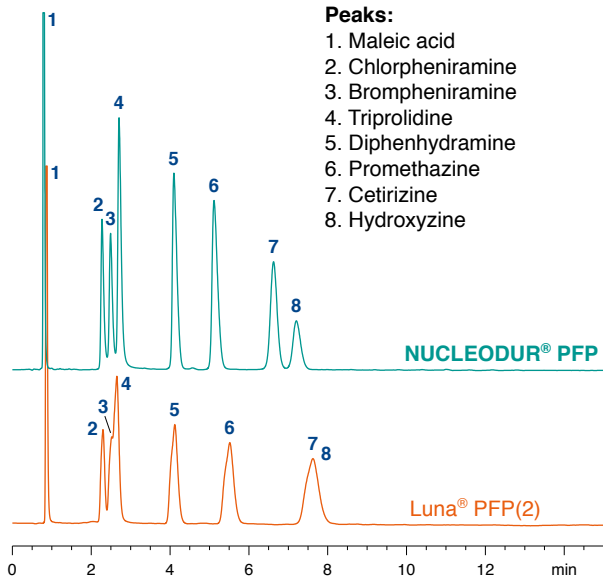
1. Ascorbic acid
2. Paracetamol
3. Caffeine
4. Chlorpheniramine
5. Dextromethorphan
6. Diphenhydramine



## Antihistamines

MN Appl. No. 124851

Columns: 100 x 4.6 mm NUCLEODUR® PFP, 5 µm  
 100 x 4.6 mm Phenomenex Luna® PFP(2), 5 µm  
 Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3.0 (30:70, v/v)  
 Flow rate: 1.3 mL/min  
 Temperature: 30 °C  
 Detection: UV, 210 nm  
 Injection volume: 1 µL



### Peaks:

1. Maleic acid
2. Chlorpheniramine
3. Brompheniramine
4. Triprolidine
5. Diphenhydramine
6. Promethazine
7. Cetirizine
8. Hydroxyzine

## β<sub>2</sub>-Agonists in human urine by LC-MS/MS

MN Appl. No. 119760

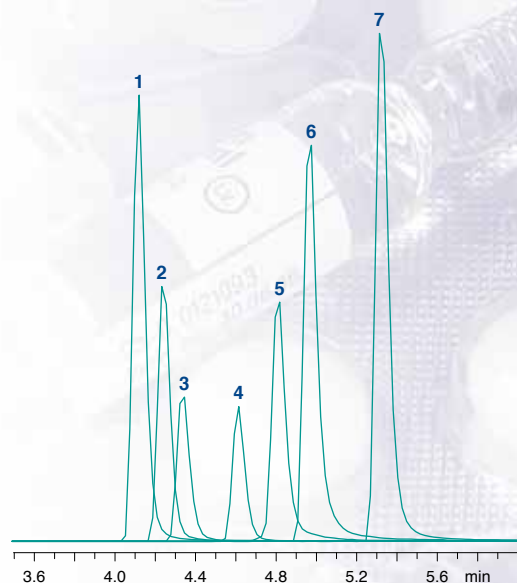
Column: 75 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
 Sample prep.: please refer to Thevis et al., J. Mass Spectrom 38 (2003) 1197–1206  
 Eluents: A) 5 mM ammonium acetate with 0.1 % acetic acid, pH 3.5; B) acetonitrile; 0 % B → 100 % B in 6 min, reequilibration at 100 % A for 3.5 min  
 Flow rate: 0.8 mL/min  
 Temperature: 25 °C  
 Detection: electrospray ionization / multiple reaction monitoring (MRM) on an Applied Biosystems API 2000  
 Injection volume: 20 µL

### LC-MS/MS chromatogram

2 mL urine aliquot fortified with 200 ng each

### Peaks:

1. Reproterol (4.12 min)
2. Fenoterol (4.24 min)
3. Ritodrine (4.34 min)
4. Ractopamine (4.61 min)
5. Clenbuterol (4.81 min)
6. Bambuterol (4.97 min)
7. Mapenterol (5.32 min)

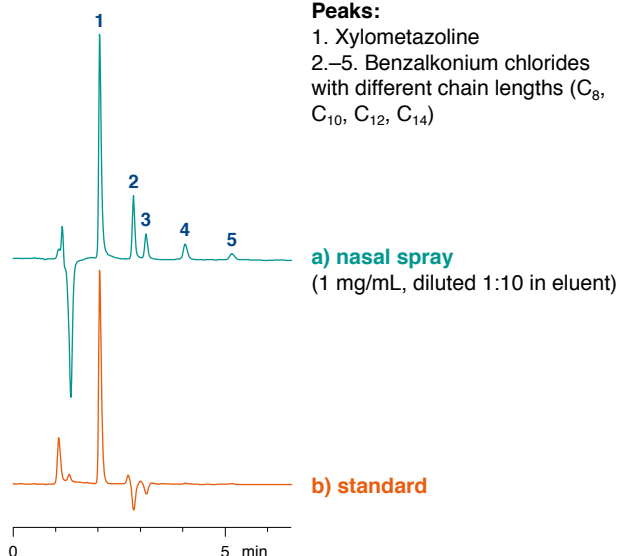


Courtesy of M. Thevis and W. Schänzer, Institute of Biochemistry, German Sport University, Cologne, Germany.

## Xylometazoline in nasal spray

MN Appl. No. 120390

Column: 125 x 4 mm NUCLEODUR® 100-5 CN-RP  
 Eluent: acetonitrile – 50 mM Na citrate, pH 3.0 (50:50, v/v)  
 Flow rate: 0.8 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm  
 Injection volume: 100 µL



### Peaks:

1. Xylometazoline
- 2.–5. Benzalkonium chlorides with different chain lengths (C<sub>8</sub>, C<sub>10</sub>, C<sub>12</sub>, C<sub>14</sub>)

### a) nasal spray

(1 mg/mL, diluted 1:10 in eluent)

### b) standard

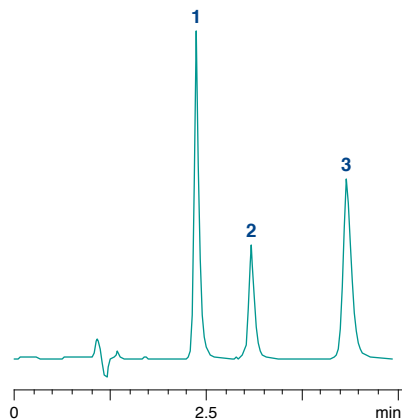
# Applications

## Basic drugs

### MN Appl. No. 119320

Column: 125 x 4 mm NUCLEODUR® 100-5 CN-RP  
Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 6.5 (50:50, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 1.0 µL

Peaks:	Tailing factor
1. Procainamide (5 ng/µL)	1.3
2. Clonidin (10 ng/µL)	1.2
3. Clenbuterol (12 ng/µL)	1.2

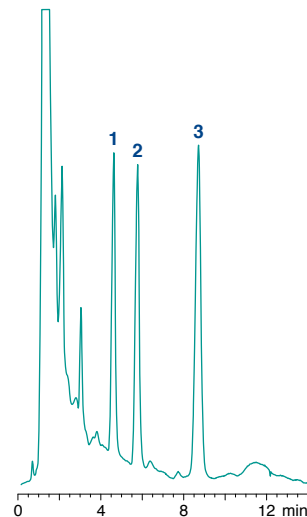


## Benzodiazepine midazolam and metabolite from plasma

### MN Appl. No. 118470

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 3 µm  
Eluent: 127 mL KH<sub>2</sub>PO<sub>4</sub> (9.1 g/L H<sub>2</sub>O) + 309 mL Na<sub>2</sub>HPO<sub>4</sub> (11.9 g/L H<sub>2</sub>O) + 852 mL methanol + 0.15 g octanesulfonic acid, pH 5.56  
Flow rate: 0.7 mL/min  
Temperature: 25 °C  
Detection: UV, DAD

- Peaks:**
1. α-Hydroxymidazolam (metabolite)
  2. Midazolam (250 ng/mL)
  3. Internal standard



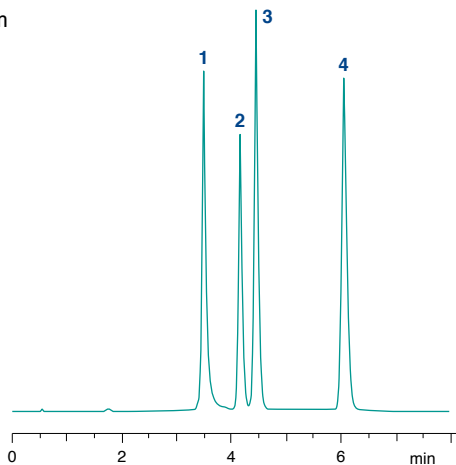
Courtesy of Mrs. Richter, Institute of Anesthetics, Biochemical Laboratory, University of Erlangen, Germany

## Benzodiazepines

### MN Appl. No. 117850

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 6.5 (45:55, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 22 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

- Peaks:**
1. Bromazepam
  2. Oxazepam
  3. Lorazepam
  4. Temazepam

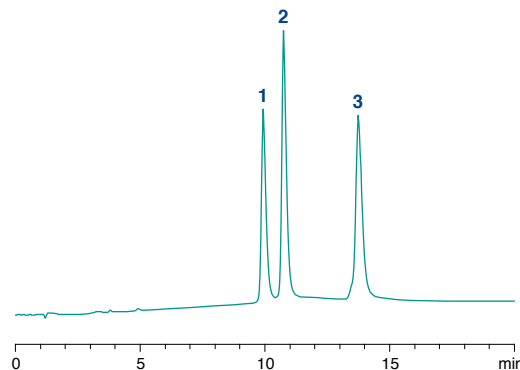


## Sedative drugs

### MN Appl. No. 119300

Column: 125 x 4 mm NUCLEODUR® 100-5 CN-RP  
Eluent: A) methanol  
B) 50 mM ammonium acetate, pH 5.0  
70% B → 50% B in 10 min (10 min)  
Flow rate: 1.5 mL/min  
Temperature: 30 °C  
Detection: UV, 254 nm  
Injection volume: 1 µL (1 + 2: 670 µg/mL, 3: 335 µg/mL)

- Peaks:**
1. Promethazine
  2. Promazine
  3. Chlorpromazine





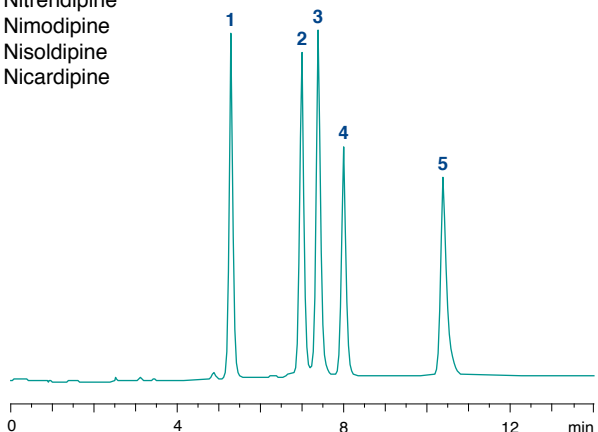
## Coronary therapeutic drugs (Ca-antagonists)

**MN Appl. No. 119310**

Column: 125 x 4 mm NUCLEODUR® 100-5 CN-RP  
 Eluent: A) acetonitrile, B) 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 6.5  
 30% B → 50% B in 7.5 min (7.5 min)  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 254 nm  
 Injection volume: 2.5 µL (25 µg/mL each)

**Peaks:**

1. Nifedipine
2. Nitrendipine
3. Nimodipine
4. Nisoldipine
5. Nicardipine



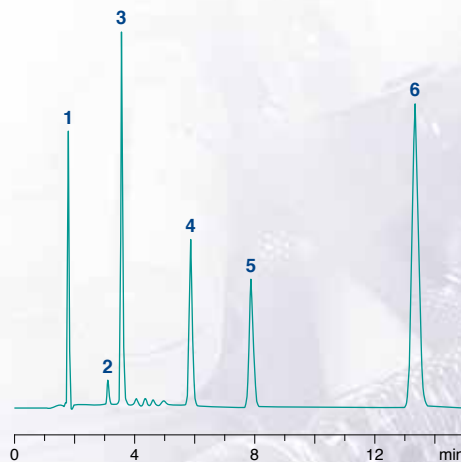
## Antibacterial drugs

**MN Appl. No. 117870**

Column: 250 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
 Eluent: acetonitrile – water (40:60, v/v) 0.05% TFA  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 254 nm  
 Injection volume: 5 µL

**Peaks:**

1. Ofloxacin
2. Ciprofloxacin
3. Cinoxacin
4. Penicillin G
5. Penicillin V
6. Cloxacillin



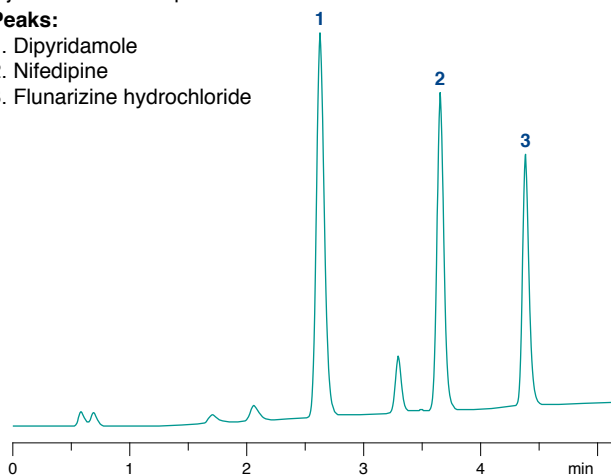
## Cardiovascular drugs

**MN Appl. No. 122560**

Column: 75 x 4.6 mm NUCLEODUR® C<sub>18</sub> Gravity, 3 µm  
 Eluent: A) 50 mM KH<sub>2</sub>PO<sub>4</sub> + Na pentanesulfonate pH 2.5  
 B) methanol  
 45% B → 90% B in 6 min  
 Flow rate: 1.3 mL/min  
 Temperature: 35 °C  
 Detection: UV, 230 nm  
 Injection volume: 5 µL

**Peaks:**

1. Dipyridamole
2. Nifedipine
3. Flunarizine hydrochloride



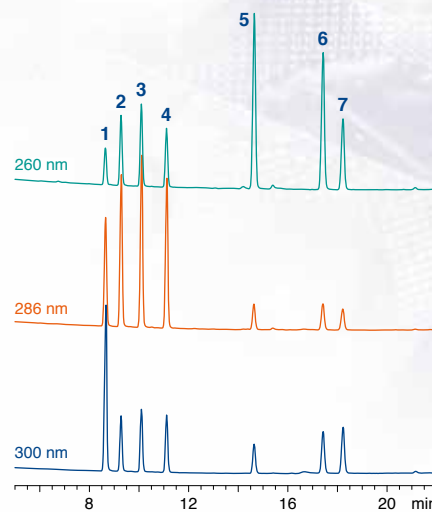
## Gyrase inhibitors

**MN Appl. No. 120400**

Column: 150 x 3 mm NUCLEODUR® Sphinx RP, 5 µm  
 Eluent: A) 0.05 M H<sub>3</sub>PO<sub>4</sub>, B) acetonitrile  
 5% B → 50% B in 20 min  
 Flow rate: 0.5 mL/min  
 Detection: UV DAD, 260 nm, 286 nm and 300 nm  
 Injection volume: 20 µL (0.625 ng/µL of each compound)

**Peaks:**

1. Marbofloxacin
2. Ciprofloxacin
3. Enrofloxacin
4. Sarafloxacin
5. Oxolinic acid
6. Nalidixic acid
7. Flumequine



Courtesy of R. Lippold, Chemical and Veterinary Research Agency, Freiburg, Germany.

# Applications

## Quinolone antibiotics

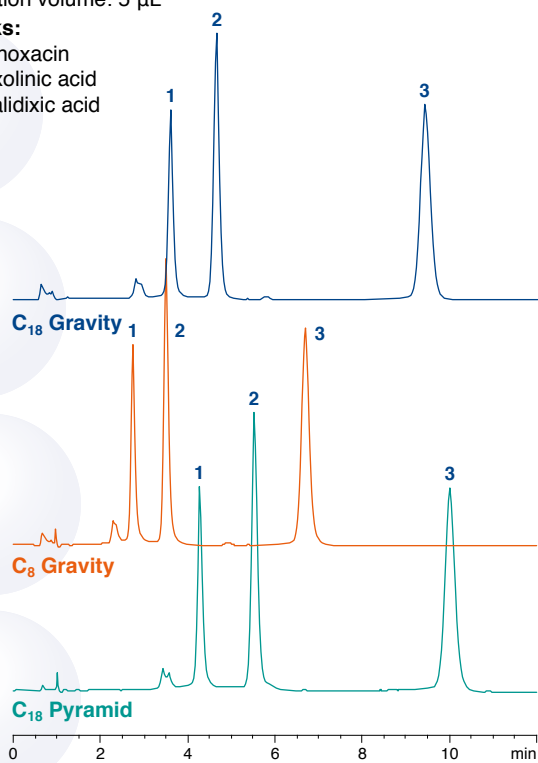
**MN Appl. No. 120460/120470**

Columns: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
125 x 4 mm NUCLEODUR® C<sub>8</sub> Gravity, 5 µm  
125 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm

Eluent: methanol – 0.2% formic acid (40:60, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 30 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

### Peaks:

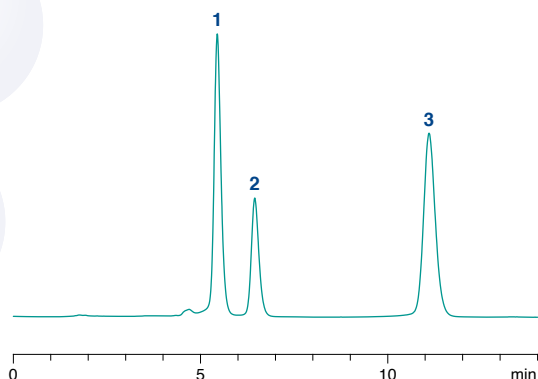
1. Cinoxacin
2. Oxolinic acid
3. Nalidixic acid



**MN Appl. No. 119870**

Column: 150 x 4.6 mm NUCLEODUR® Sphinx RP, 5 µm  
Eluent: methanol – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 2.5 (50:50, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 22 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

### Peaks as listed above



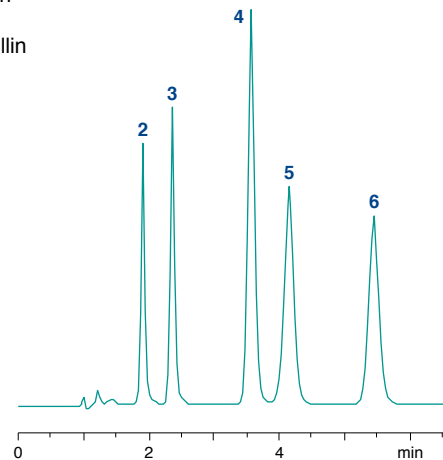
## Penicillin antibiotics

**MN Appl. No. 117860**

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
Eluent: acetonitrile – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3.0 (40:60, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 22 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

### Peaks:

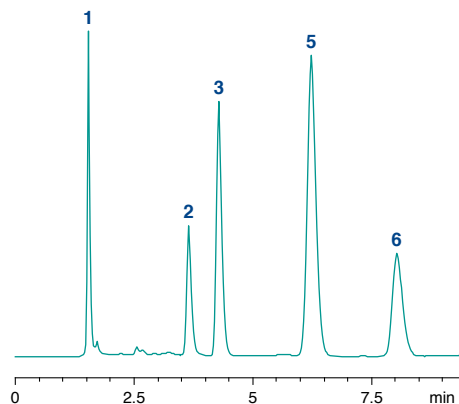
1. Amoxicillin
2. Penicillin G
3. Penicillin V
4. Cloxacillin
5. Nafcillin
6. Dicloxacillin



**MN Appl. No. 119150**

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
Eluent: acetonitrile – 0.1% TFA (50:50, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 1 µL

### Peaks as listed above



## $\beta$ -lactam antibiotics

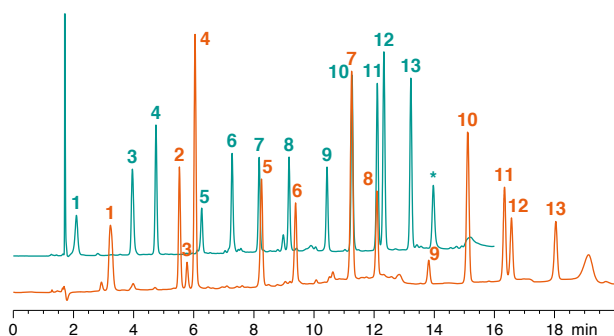
MN Appl. No. 124840

Column: 150 x 3 mm NUCLEODUR® PFP, 5  $\mu$ m  
 Eluents: A) acetonitrile, B) 25 mM  $\text{KH}_2\text{PO}_4$ , pH 2.7  
 10% A  $\rightarrow$  60% A in 15 min

Flow rate: 0.563 mL/min  
 Temperature: 40  $^\circ\text{C}$ , 50  $^\circ\text{C}$   
 Detection: UV, 220 nm, UV, 240 nm  
 Injection volume: 1  $\mu$ L

### Peaks:

- |                |                 |                   |
|----------------|-----------------|-------------------|
| 1. Amoxicillin | 6. Cefamandole  | 11. Cloxacillin   |
| 2. Cephalexin  | 7. Cefalotin    | 12. Nafcillin     |
| 3. Ampicillin  | 8. Piperacillin | 13. Dicloxacillin |
| 4. Cefotaxime  | 9. Penicillin V | * Impurity        |
| 5. Cefoxitin   | 10. Oxacillin   |                   |



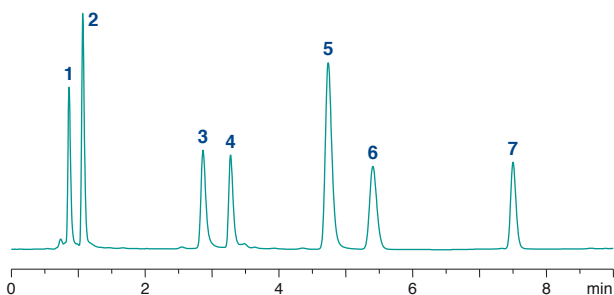
MN Appl. No. 123760

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> HTec, 5  $\mu$ m  
 Eluent: A) acetonitrile, B) 0.05% TFA in water  
 70% B (1 min)  $\rightarrow$  60% B in 0.5 min (3.5 min)  $\rightarrow$   
 50% B in 1 min  $\rightarrow$  37.5% B in 4 min

Flow rate: 0.9 mL/min  
 Temperature: 25  $^\circ\text{C}$   
 Detection: UV, 254 nm  
 Injection volume: 10  $\mu$ L  
 Concentration: 300  $\mu\text{g/mL}$

### Peaks:

1. Amoxicillin
2. Enrofloxacin
3. Cinoxacin
4. Oxolinic acid
5. Nalidixic acid
6. Penicillin V
7. Cloxacillin



## Anticoccidial drugs (polyether antibiotics)

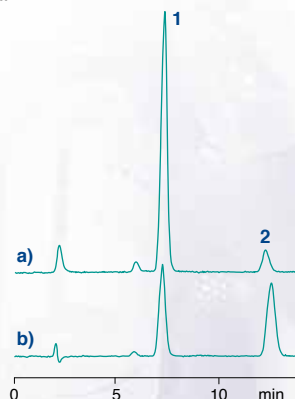
MN Appl. No. 118760

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 3  $\mu$ m  
 Eluent: methanol – 50 mmol phosphate buffer pH 3.0 –  
 methylheptylamine (900:99:1, v/v/v)

Flow rate: 0.7 mL/min  
 Temperature: 23  $^\circ\text{C}$   
 Detection: UV/VIS, 600 nm after post column derivatization  
 with dimethylaminobenzaldehyde (0.4 mL/min)  
 Injection volume: 100  $\mu$ L

### Peaks:

1. Monensin sodium
2. Salinomycin sodium



- a): Sample spiked with monensin sodium and content of salinomycin sodium  
 b): Standard of monensin sodium and salinomycin sodium

Courtesy of J. Schönherr, Saxon State Institute for Agriculture, Leipzig, Germany

## Cephalosporin antibiotics

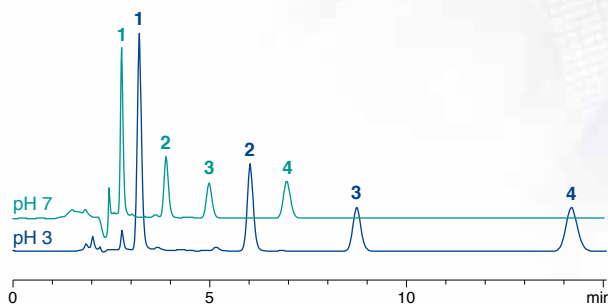
MN Appl. No. 122580/122590

Column: 150 x 4.6 mm NUCLEODUR® C<sub>18</sub> Gravity, 5  $\mu$ m  
 Eluent: acetonitrile – 25 mM  $\text{KH}_2\text{PO}_4$  (20:80, v/v)  
 pH 3 with  $\text{H}_3\text{PO}_4$ , pH 7

Flow rate: 0.8 mL/min  
 Temperature: 35  $^\circ\text{C}$   
 Detection: UV, 254 nm  
 Injection volume: 2  $\mu$ L

### Peaks:

1. Cefotaxime
2. Cefoxitin
3. Cefamandole
4. Cephalothin



Protonation causes a drastic increase in retention time, but an improved peak symmetry.

# Applications

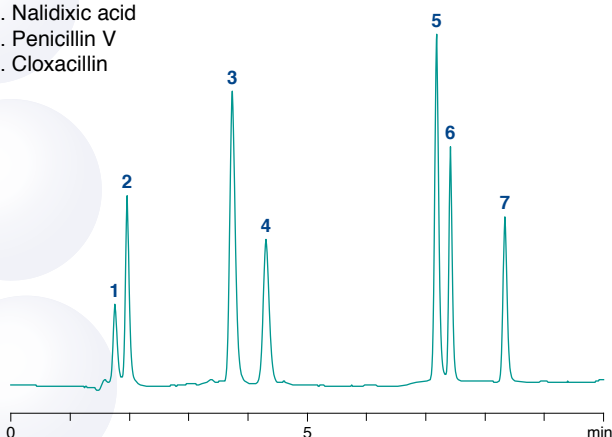
## Antibacterial drugs

### MN Appl. No. 122470

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluent: A) acetonitrile, B) water + 0.05 % TFA  
60 % B (4 min) → 40 % B in 1 min (5 min)  
Flow rate: 0.9 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

#### Peaks:

1. Amoxicillin
2. Enrofloxacin
3. Cinoxacin
4. Oxolinic acid
5. Nalidixic acid
6. Penicillin V
7. Cloxacillin



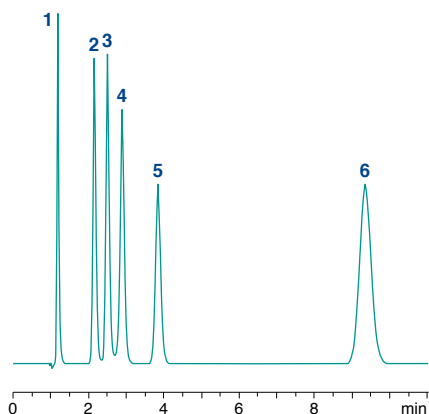
## Sulfonamides

### MN Appl. No. 117880

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluent: methanol – 0.1 % TFA (20:80, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 22 °C  
Detection: UV, 230 nm  
Injection volume: 4 µL

#### Peaks:

1. Sulfanilamide
2. Sulfadiazine
3. Sulfathiazole
4. Sulfamerazine
5. Sulfadimidine
6. Succinylsulfathiazole



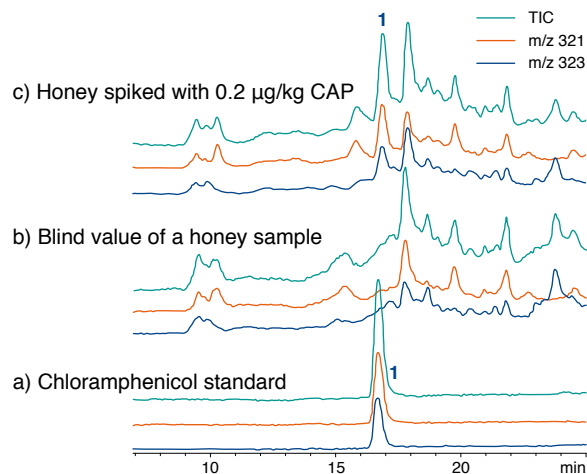
## Determination of chloramphenicol residues in honey by microbore HPLC

### MN Appl. No. 119810

Column: 100 x 1 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluent: A) methanol, B) water  
15 % A → 80 % A in 9 min (15 min) → 15 % A in 1 min; injection after 7 min  
Flow rate: 60 µL/min  
Detection: MS  
Injection volume: 1 µL

#### Peaks

1. Chloramphenicol (CAP)



S. Oepkemeier, H.D. Winkler, GIT 46 (2002) 982–985.

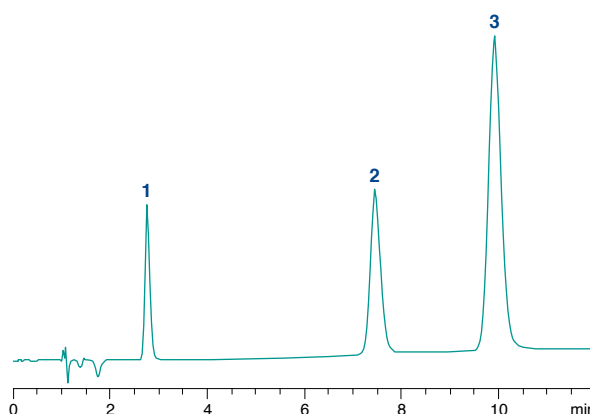
## Separation of theobromine, vanillin and caffeine

### MN Appl. No. 119920

Column: 125 x 4 mm NUCLEODUR® Sphinx RP, 5 µm  
Eluent: methanol – 1.25 % acetic acid (20:80, v/v)  
Flow rate: 1 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 0.8 µL

#### Peaks:

1. Theobromine
2. Caffeine
3. Vanillin

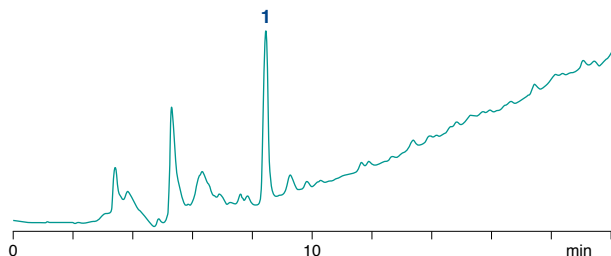


## Alkaline tannic acid mixture

MN Appl. No. 120450

Column: 250 x 4.6 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
 Eluent: A) 0.425 % H<sub>3</sub>PO<sub>4</sub>, pH 1.4, B) acetonitrile  
 5% B → 25% B in 15 min (5 min) → 5% B in  
 2 min (3 min)  
 Flow rate: 0.8 mL/min  
 Detection: UV, 275 nm (optimized for gallic acid)  
 Injection volume: 10 µL

**Peaks:**  
 1. Gallic acid



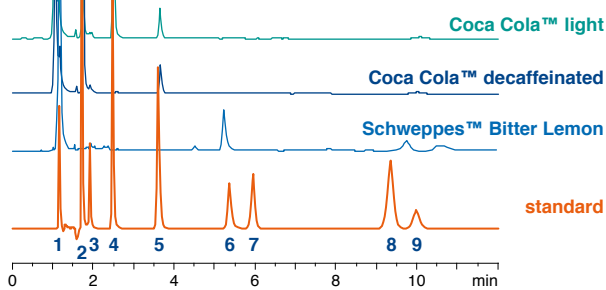
## Soft drink additives

MN Appl. No. 118560

Column: 150 x 4.6 mm NUCLEODUR® 100-5 C<sub>8</sub> ec  
 Eluent: 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3 – acetonitrile (5:1, v/v)  
 Flow rate: 1.9 mL/min  
 Temperature: 25 °C  
 Detection: UV, 220 nm  
 Injection volume: 10 µL

**Peaks:**  
 1. Ascorbic acid (acidic)  
 2. Acesulfame K  
 3. Saccharin  
 4. Caffeine (basic)  
 5. Aspartame  
 6. Quinine (strongly basic)  
 7. Vanillin  
 8. Sorbic acid (acidic)  
 9. Benzoic acid (acidic)

Soft drink samples were degassed for 5 min and injected undiluted.



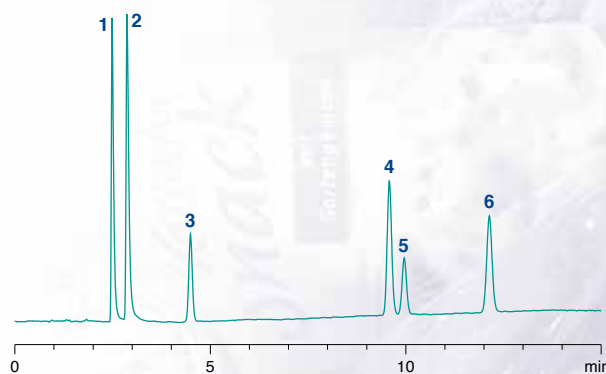
For fast separation of sweeteners on NUCLEODUR® 100-5 C<sub>18</sub> ec see appl. 117940 at [www.mn-net.com](http://www.mn-net.com).

## Sweeteners

MN Appl. No. 123750

Column: 150 x 4.6 mm NUCLEODUR® C<sub>18</sub> HTec, 5 µm  
 Eluent: A) acetonitrile, B) 25 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3.5  
 15% A (2.5 min) → 25% A in 9.5 min (3 min)  
 Flow rate: 1.3 mL/min  
 Temperature: 40 °C  
 Detection: UV, 220 nm  
 Injection volume: 5 µL  
 Concentration: 0.1 mg/mL each

**Peaks:**  
 1. Acesulfame K  
 2. Saccharin  
 3. Aspartame  
 4. Benzoic acid  
 5. Sorbic acid  
 6. Dehydroacetic acid

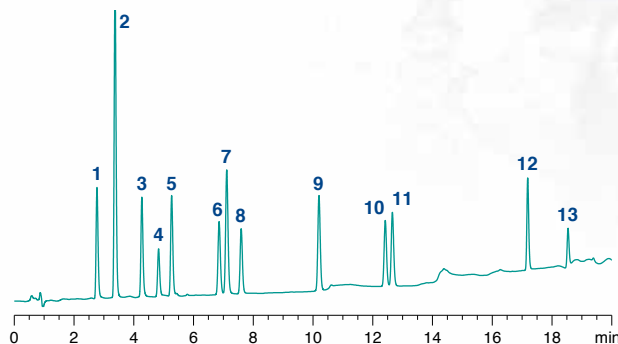


## Preservatives

MN Appl. No. 124590

Column: 150 x 3 mm NUCLEODUR® PolarTec, 3 µm  
 Eluent: A) acetonitrile, 0.1 % TFA, B) water, 0.1 % TFA  
 20% A → 50% A in 12 min → 65% A in 2 min →  
 95% A in 6 min  
 Flow rate: 0.9 mL/min  
 Temperature: 45 °C  
 Detection: UV, 220 nm  
 Injection volume: 5 µL

**Peaks:**  
 1. Benzyl alcohol  
 2. Phenoxyethanol  
 3. Dehydroacetic acid  
 4. *p*-Anisic acid  
 5. Methyl paraben  
 6. Salicylic acid  
 7. Benzoic acid  
 8. Ethyl paraben  
 9. Propyl paraben  
 10. Isobutyl paraben  
 11. Butyl paraben  
 12. Irgasan  
 13. 3,3,4-Triclocarbanilid



# Applications

## Food dyes

### MN Appl. No. 122500/122510

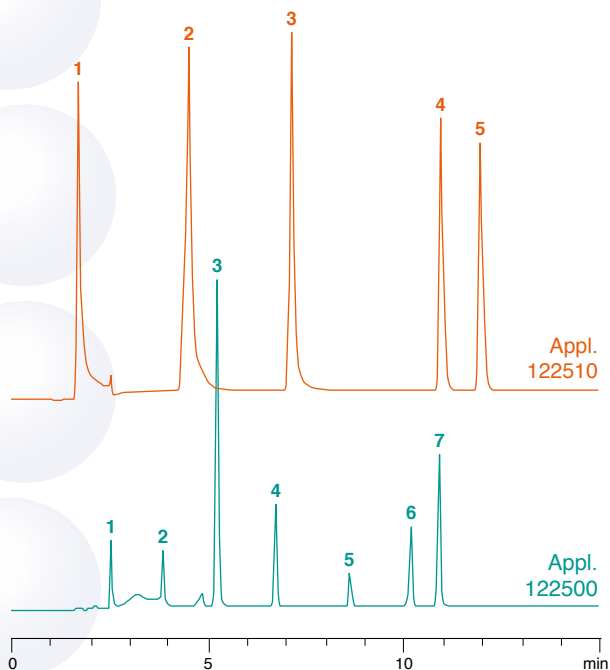
Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluent: A) acetonitrile, B) 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 5  
95% B → 50% B in 20 min → 20% B in 5 min  
→ 95% B in 1 min (4 min)  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

#### Peaks application 122510

1. Ponceau 6R (E 126)
2. Ponceau 4R (E 124)
3. Azorubine (E 122)
4. Erythrosine (E 127)
5. Fast Red E

#### Peaks application 122500

- 1., 2. Tartrazine (E 102)
3. Fast Yellow
- 4.-6. Quinoline yellow (E 104)
7. Yellow orange S (sunset yellow CFC, E 110)



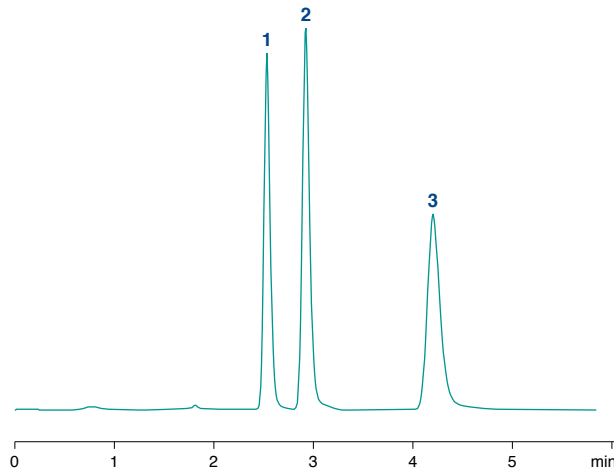
## Acrylamide, methacrylamide and methacrylic acid

### MN Appl. No. 123010

Column: 125 x 4 mm NUCLEODUR® HILIC, 5 µm  
Eluent: acetonitrile – 0.1% formic acid (98:2, v/v)  
Flow rate: 0.6 mL/min  
Temperature: 22 °C  
Detection: UV, 210 nm  
Injection volume: 0.5 µL

#### Peaks:

1. Methacrylamide
2. Acrylamide
3. Methacrylic acid



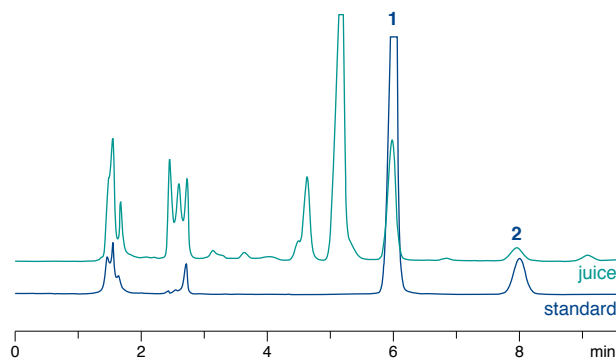
## Patulin and hydroxymethylfurfural in apple juice

### MN Appl. No. 121800

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
+ 8 x 4 mm guard column  
Sample prep.: see appl. 121800 at [www.mn-net.com](http://www.mn-net.com)  
Eluent: water – acetonitrile (95:5, v/v)  
Flow rate: 1.5 mL/min  
Detection: UV, 276 nm  
Injection volume: 10 µL

#### Peaks:

1. Hydroxymethylfurfural
2. Patulin



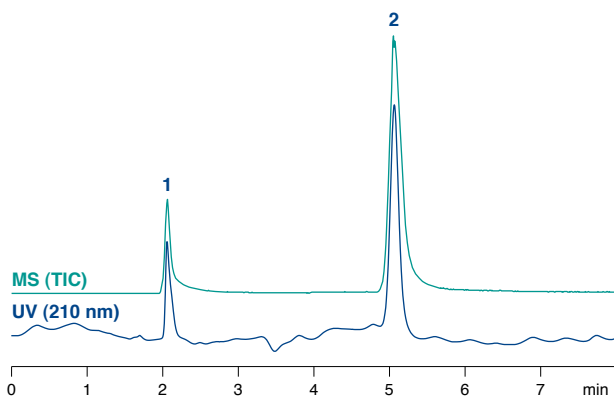
Courtesy of A. Gessler, Wesergold Getränkeindustrie GmbH & Co. KG, Rinteln, Germany.

## Melamine and cyanuric acid

**MN Appl. No. 123070**

Column: 125 x 2 mm NUCLEODUR® HILIC, 5 µm  
 Eluent: acetonitrile – 10 mM ammonium formate, pH 4 (90:10, v/v)  
 Flow rate: 0.2 mL/min  
 Temperature: 25 °C  
 Detection: UV, 210 nm and MS (TIC)  
 Injection volume: 2 µL

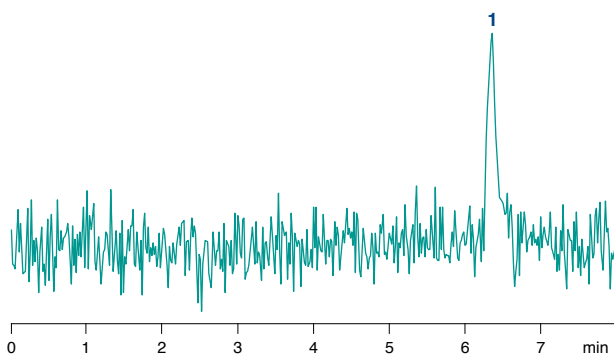
**Peaks:**  
 1. Melamine (10 µg/mL)  
 2. Cyanuric acid (490 µg/mL)



**MN Appl. No. 123090**

Column: 125 x 4 mm NUCLEODUR® HILIC, 5 µm  
 Eluent: acetonitrile – 10 mM ammonium formate, pH 4 (90:10, v/v)  
 Flow rate: 0.6 mL/min  
 Temperature: 25 °C  
 Detection: MS  
 Injection volume: 1 µL

**Peaks:**  
 1. Melamine in milk (100 pg/injection)

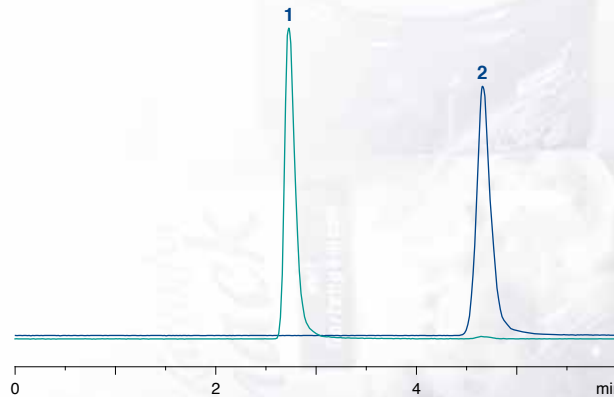


## Creatinine and creatine

**MN Appl. No. 123000**

Column: 125 x 2 mm NUCLEODUR® HILIC, 3 µm  
 Eluent: acetonitrile – 10 mM ammonium acetate, pH 4 (70:30, v/v)  
 Flow rate: 0.2 mL/min  
 Temperature: 25 °C  
 Detection: MS  
 Injection volume: 5 µL (30 ng/µL)

**Peaks:**  
 1. Creatinine  
 2. Creatine



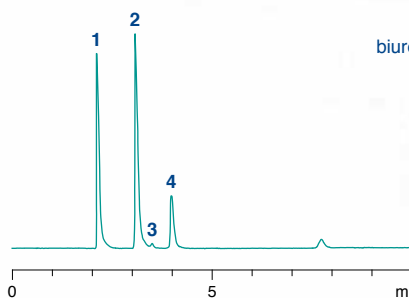
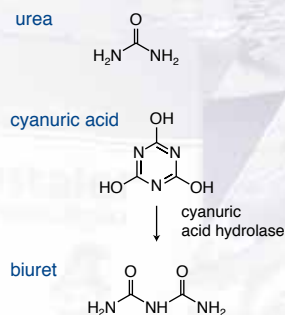
For UV detection see appl. 122990 at [www.mn-net.com](http://www.mn-net.com).

## Separation of urea, biuret and cyanuric acid

**MN Appl. No. 120440**

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
 Eluent: water (100%)  
 Flow rate: 1 mL/min  
 Detection: UV, 190 nm  
 Injection volume: 10 µL

**Peaks:**  
 1. Urea (0.5 mg/mL)  
 2. Biuret (0.09 mg/mL)  
 3. impurity in biuret  
 4. Cyanuric acid (0.05 mg/mL)



Courtesy of C. Greve, Institute of Chemical Engineering, University of Clausthal, Germany.

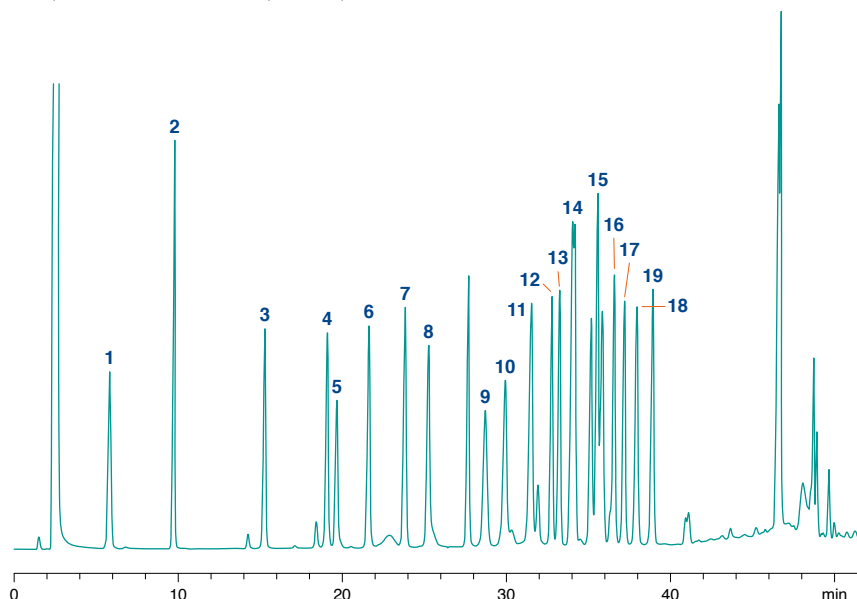
## Amino acids as OPA derivatives

MN Appl. No. 118450

Column: 250 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
Eluent: A) methanol – acetonitrile (50:50, v/v), B) Na acetate buffer pH 6.5 + 5 % A)  
100 % B (2.9 min) → 95 % B in 3.1 min → 85 % B in 11 min → 83 % B in 3 min → 70 % B in 10 min → 62 % B in 8 min → 35 % B in 7 min → 0 % B in 1 min (2 min) → 80 % B in 0.5 min (2.5 min) → 100 % B in 0.1 min  
Flow rate: 1 mL/min  
Detection: fluorescence,  
 $\lambda_{\text{ex}}$  230 nm,  $\lambda_{\text{em}}$  450 nm

### Peaks:

1. Aspartic acid
2. Glutamic acid
3. Asparagine
4. Serine
5. Glutamine
6. Histidine
7. Glycine
8. Alanine
9. Arginine
10.  $\gamma$ -Aminobutyric acid
11. Tyrosine
12. Valine
13. Methionine
14. Norvaline (int. std.)
15. Tryptophan
16. Phenylalanine
17. Isoleucine
18. Leucine
19. Lysine



Courtesy of Mr. Zürcher, Technical University of Munich, Chair of Brewing Technology, Freising Weihenstephan, Germany.  
For separation of amino acids also see appl. 120510 at [www.mn-net.com](http://www.mn-net.com).

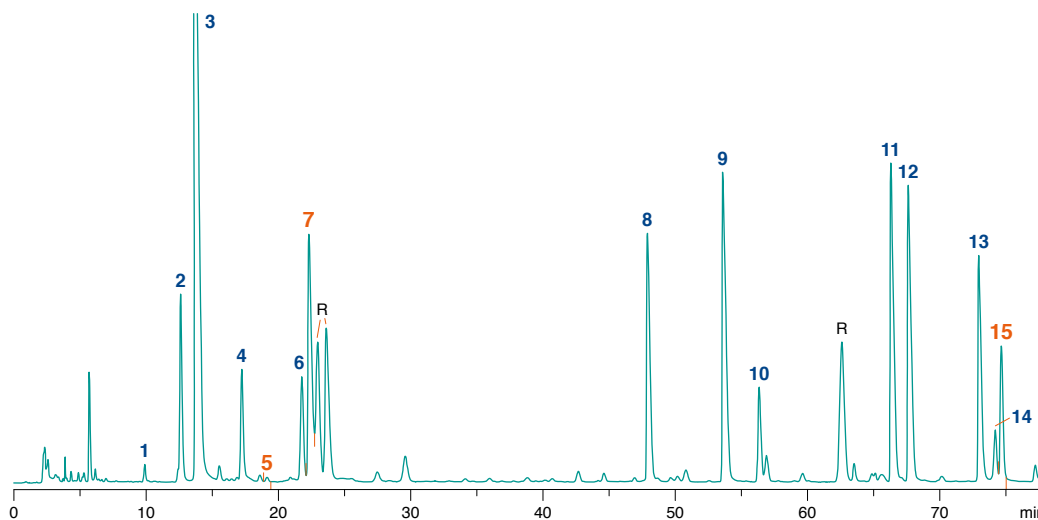
## Determination of physiological amino acids from supernatants of cell cultures

MN Appl. No. 118980

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 3  $\mu$ m  
Sample preparation: supernatants from cell cultures are deproteinated, derivatized with phenylisothiocyanate and filtered  
Eluent: A) 70 mM sodium acetate, pH 6.5, 2.5 % acetonitrile, 1 ppm EDTA; B) acetonitrile – water – methanol (45:40:15, v/v/v)  
3 % B (2 min) → 7.5 % B in 16 min (8 min) → 44 % B in 49 min  
(washing: 100 % B for 10 min; equilibration: 3 % B for 5 min)  
Flow rate: 0.8 mL/min  
Temperature: 47  $\pm$  1 °C  
Detection: UV, 254 nm  
Injection volume: 40  $\mu$ L

### Peaks:

1. 4-Hydroxyproline
  2. Serine
  3. Glutamine
  4. Histidine
  5. Citrulline (5.90  $\mu$ mol/L)
  6. Threonine
  7. Arginine (504.33  $\mu$ mol/L)
  8. Tyrosine
  9. Valine
  10. Methionine
  11. Isoleucine
  12. Leucine
  13. Phenylalanine
  14. Tryptophan
  15. Ornithine (143.54  $\mu$ mol/L)
- R = reagents



Citrulline, arginine and ornithine were determined quantitatively.

Courtesy of Dr. J. Weinreich, Center for Medical Research, Clinic for General Surgery, University Clinical Center, Tübingen, Germany.



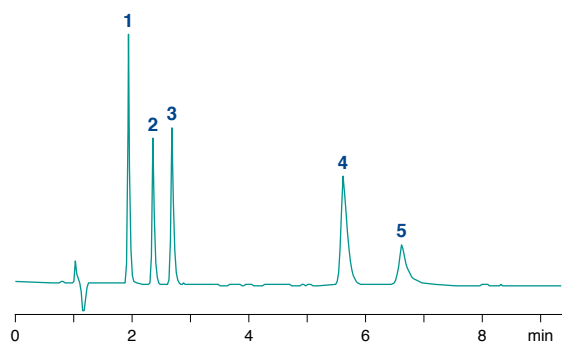
## Aromatic amino acids and histamine

**MN Appl. No. 122980**

Column: 125 x 4 mm NUCLEODUR® HILIC, 3 µm  
 Eluent: acetonitrile – 100 mM ammonium acetate, pH 4 (75:25, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 218 nm  
 Injection volume: 0.5 µL

### Peaks:

1. Phenylalanine
2. Phenylglycine
3. Tyrosine
4. Histamine
5. Histidine



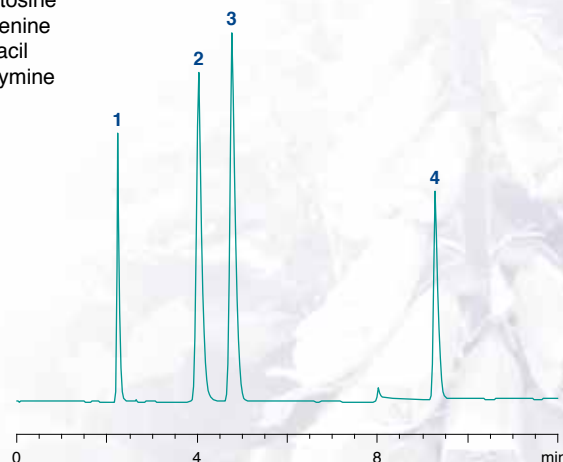
## Nucleic acid bases

**MN Appl. No. 119140**

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
 Eluent: A) 50 mM NH<sub>4</sub>H<sub>2</sub>PO<sub>4</sub>, pH 2.5, B) acetonitrile 100% A (2.5 min) → 90% A in 10 min  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 254 nm  
 Injection volume: 3 µL

### Peaks:

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



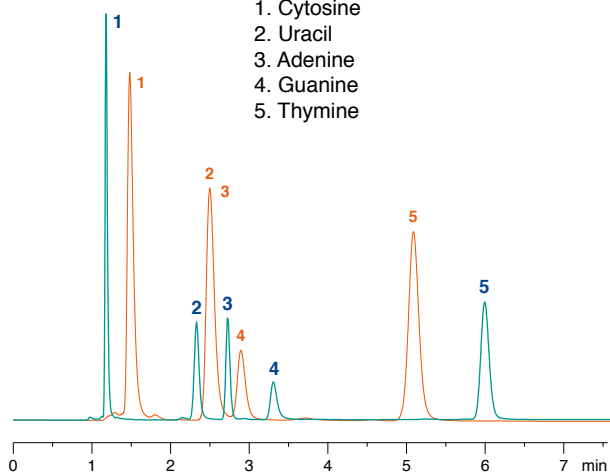
## Nucleic acid bases

**MN Appl. No. 124672**

Columns: 150 x 3 mm NUCLEODUR® PolarTec, 5 µm  
 150 x 3 mm Waters SymmetryShield™ RP18, 5 µm  
 Eluent: 30 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3.0  
 Flow rate: 0.5 mL/min  
 Temperature: 30 °C  
 Detection: UV, 220 nm  
 Injection volume: 1 µL, 3 µL

### Peaks:

1. Cytosine
2. Uracil
3. Adenine
4. Guanine
5. Thymine

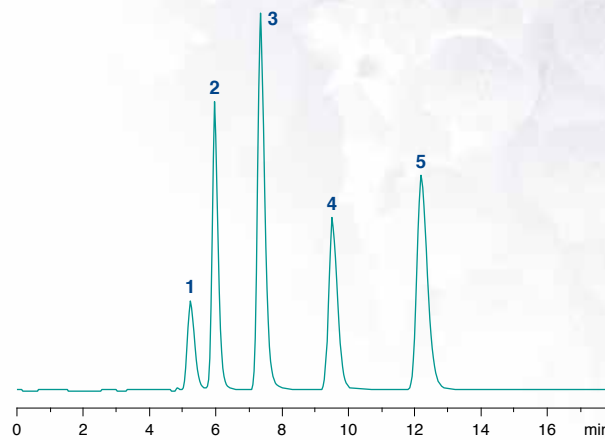


**MN Appl. No. 122950**

Column: 125 x 4 mm NUCLEODUR® HILIC, 5 µm  
 Eluent: acetonitrile – 5 mM ammonium acetate (80:20, v/v)  
 Flow rate: 0.3 mL/min  
 Temperature: 25 °C  
 Detection: UV, 254 nm

### Peaks:

1. Thymine
2. Uracil
3. Adenine
4. Cytosine
5. Guanosine



Separation of all five nucleic acid bases just on NUCLEODUR® PolarTec

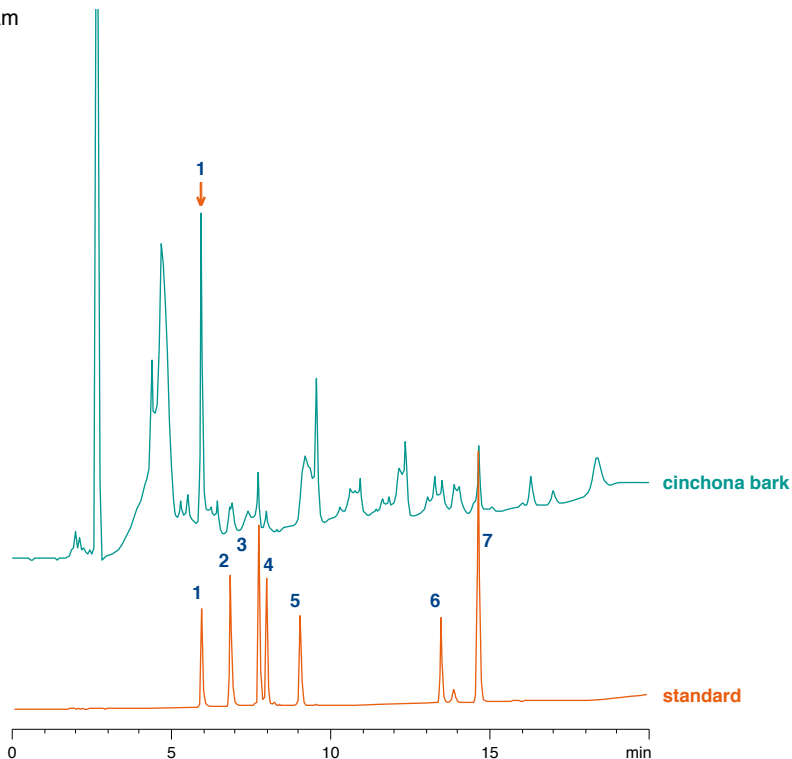
## Determination of quinine in cinchona bark

MN Appl. No. 118580

Column: 125 x 4 mm NUCLEODUR® C<sub>8</sub> Gravity, 5 µm  
 Eluent: A) 20 mM NH<sub>4</sub>H<sub>2</sub>PO<sub>4</sub>, pH 2  
 B) acetonitrile  
 Gradient: 10% B → 30% B in 15 min  
 Flow rate: 1 mL/min  
 Temperature: 30 °C  
 Detection: UV, 210 nm

**Peaks:**

1. Quinine
2. Scopolamine
3. Brucine
4. Strychnine
5. Atropine
6. Papaverine
7. Noscapine



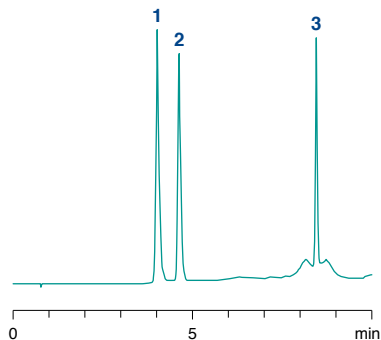
## Quinine alkaloids

MN Appl. No. 117960

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
 Eluent: A) methanol, B) 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 2.5  
 90% B → 70% B in 4 min → 30% B in 7 min  
 Flow rate: 1.3 mL/min  
 Temperature: 25 °C  
 Detection: UV, 240 nm  
 Injection volume: 10 µL

**Peaks:**

1. Chloroquine
2. Quinine
3. Mefloquine



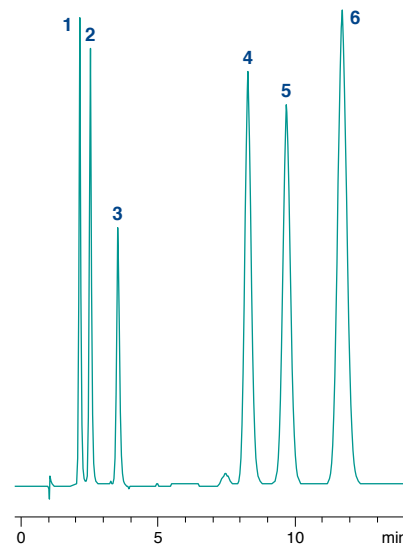
## Steroids

MN Appl. No. 118540

Column: 125 x 4 mm NUCLEODUR® C<sub>8</sub> Gravity, 5 µm  
 Eluent: acetonitrile – water (60:40, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 240 nm

**Peaks:**

1. Cortisone
2. Hydrocortisone
3. Hydrocortisone 21-acetate
4. 6α-Methyl-11β-hydroxyprogesterone
5. 6α-Methyl-17α-hydroxyprogesterone
6. 6α-Methyl-17α-hydroxyprogesterone acetate



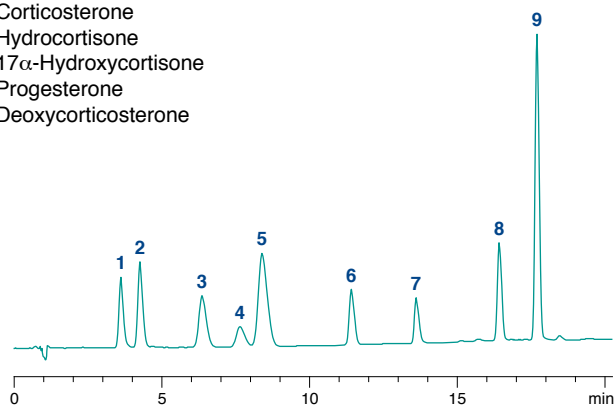
## Steroids

**MN Appl. No. 122530**

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 μm  
 Eluent: A) acetonitrile, B) water  
 70% B (7 min) → 20% B in 16 min → 70% B in  
 2 min  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 240 nm  
 Injection volume: 3 μL

**Peaks:**

1. Cortisone
2. Prednisolone
3. 6α-Methylprednisolone
4. Dexamethasone
5. Corticosterone
6. Hydrocortisone
7. 17α-Hydroxycortisone
8. Progesterone
9. Deoxycorticosterone



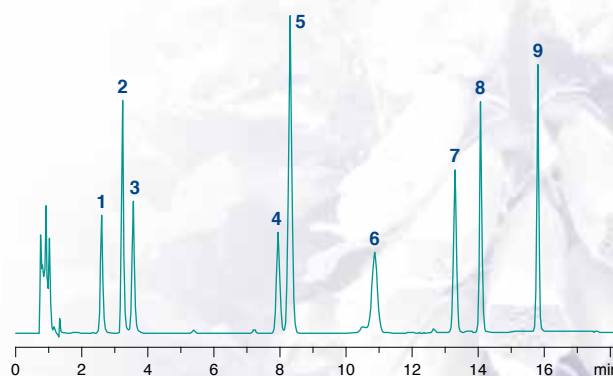
## Steroids

**MN Appl. No. 123710**

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> HTec, 5 μm  
 Eluent: A) acetonitrile, B) water  
 70% B → 60% B in 5 min (5 min) → 35% B in  
 5 min (5 min)  
 Flow rate: 1.0 mL/min  
 Temperature: 35 °C  
 Detection: UV, 254 nm  
 Injection volume: 10 μL

**Peaks:**

- |                 |                                      |
|-----------------|--------------------------------------|
| 1. Estriol      | 6. Estrone                           |
| 2. Prednisolone | 7. 6α-Methyl-11β-hydroxyprogesterone |
| 3. Cortisone    | 8. 6α-Methyl-17α-hydroxyprogesterone |
| 4. Estradiol    | 9. Progesterone                      |
| 5. Testosterone |                                      |



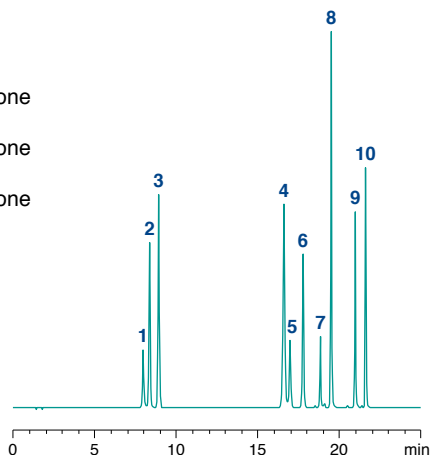
## Steroids

**MN Appl. No. 118550**

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>8</sub> ec  
 Eluent: A) water, B) methanol  
 20% B (1 min) → 35% B in 10 min (3 min) →  
 60% B in 6 min (5 min)  
 Flow rate: 1.0 mL/min  
 Temperature: 30 °C  
 Detection: UV, 230 nm  
 Injection volume: 10 μL (each ~10–50 μg/mL)

**Peaks:**

1. Estriol
2. Prednisolone
3. Cortisone
4. Testosterone
5. 6α-Methyl-11β-hydroxyprogesterone
6. 6α-Methyl-17α-hydroxyprogesterone
7. 6α-Methyl-17α-hydroxyprogesterone acetate
8. Estradiol
9. Estrone
10. Progesterone



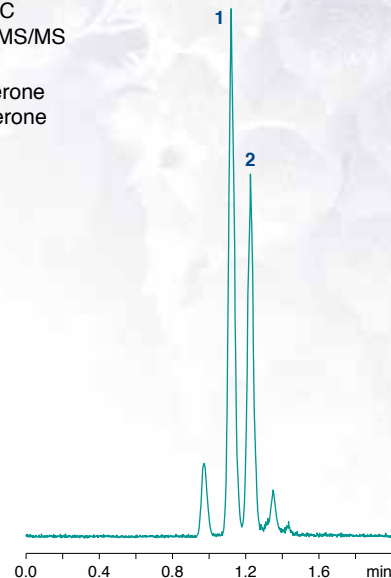
## Hydroxytestosterones from cytochrome P450

**MN Appl. No. 122140**

Column: 50 x 2 mm NUCLEODUR® C<sub>18</sub> Isis, 1.8 μm  
 Eluent: A) water + 0.1% formic acid; B) acetonitrile –  
 methanol + 0.3% formic acid  
 75% A → 60% A in 1.1 min → 0% A in 0.05 min,  
 → 75% A in 0.05 min (0.95 min)  
 Flow rate: 0.9 mL/min  
 Temperature: 70 °C  
 Detection: LC-MS/MS

**Peaks:**

1. 6β-Hydroxytestosterone
2. 7α-Hydroxytestosterone



# Applications

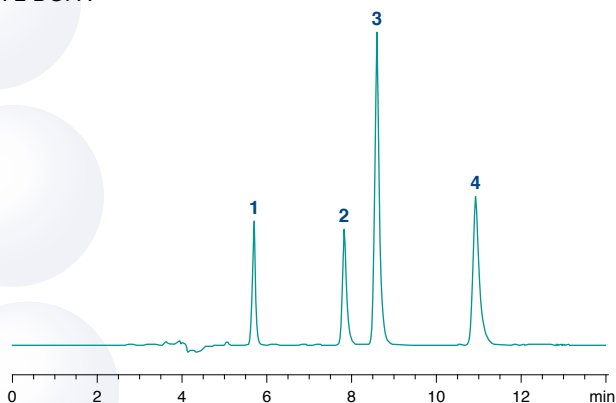
## Catecholamines

**MN Appl. No. 123030**

Column: 250 x 4 mm NUCLEODUR® HILIC, 3 µm  
Eluent: acetonitrile – 25 mM ammonium formate, pH 3 (75:25, v/v)  
Flow rate: 0.8 mL/min  
Temperature: 25 °C  
Detection: UV, 218 nm  
Injection: 5 µL, 30 ng/µL

### Peaks:

1. Norephedrine
2. Dopamine
3. Adrenaline
4. L-DOPA



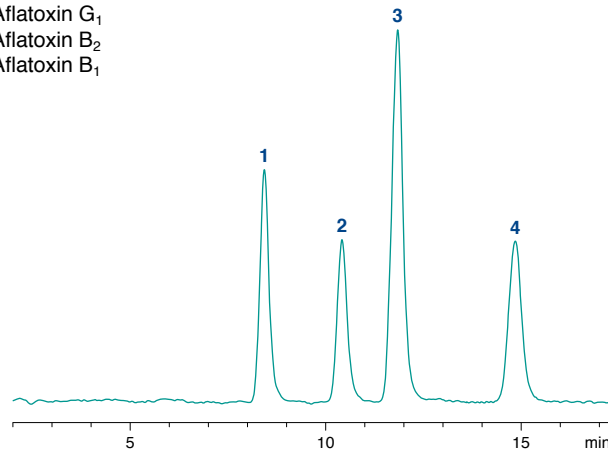
## Analysis of aflatoxins from baby food

**MN Appl. No. 120780**

Column: 250 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
Eluent: methanol – acetonitrile – water (26:17:57, v/v/v) with 119 mg KBr and 100 µL HNO<sub>3</sub> (65%) per liter  
Flow rate: 1.0 mL/min  
Detection: fluorescence, λ<sub>ex</sub> 362 nm, λ<sub>em</sub> 440 nm, post column derivatization in a CoBrA cell (Dr. Weber Consulting Kft)  
Injection volume: 100 µL

### Peaks:

1. Aflatoxin G<sub>2</sub>
2. Aflatoxin G<sub>1</sub>
3. Aflatoxin B<sub>2</sub>
4. Aflatoxin B<sub>1</sub>



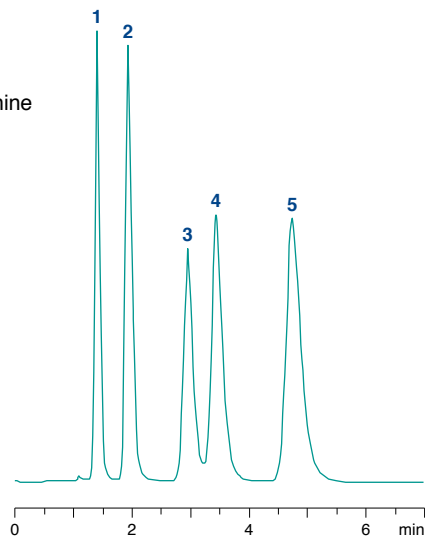
## Catecholamines

**MN Appl. No. 117930**

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluent: 100 mM NaH<sub>2</sub>PO<sub>4</sub>, pH 3.0  
Flow rate: 0.8 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

### Peaks:

1. Norephedrine
2. Adrenaline
3. Dihydroxyphenylalanine
4. Hydroxytyramine
5. Tyrosine



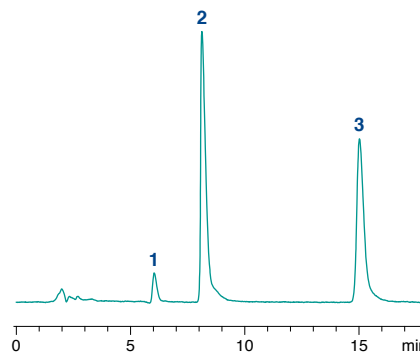
## Analysis of mycotoxins

**MN Appl. No. 119800**

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Guard column: 8 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluent: acetonitrile – water (45:55, v/v), 2 mL conc. H<sub>3</sub>PO<sub>4</sub>/L, adjusted to pH 2.6 with NaOH  
Flow rate: 0.9 mL/min  
Detection: fluorescence 273 nm and 455 nm  
Injection volume: 40 µL (7.5 ng of each substance)

### Peaks:

1. β-Zearalenol
2. α-Zearalenol
3. Zearalenone



Courtesy of K.H. Ueberschär, Federal Agricultural Research Centre, Institute of Animal Feed, Celle, Germany.

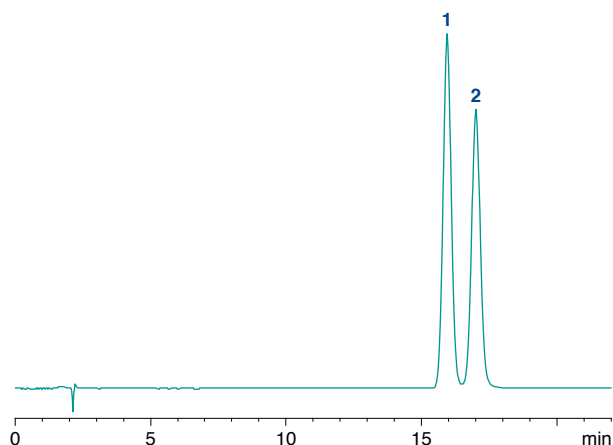
## Dexa- and betamethasone

**MN Appl. No. 121170**

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Isis, 5 µm  
 Eluent: acetonitrile – water (30:70, v/v)  
 Flow rate: 1 mL/min  
 Temperature: 25 °C  
 Detection: UV, 260 nm  
 Injection volume: 5 µL

**Peaks:**

1. Betamethasone
2. Dexamethasone



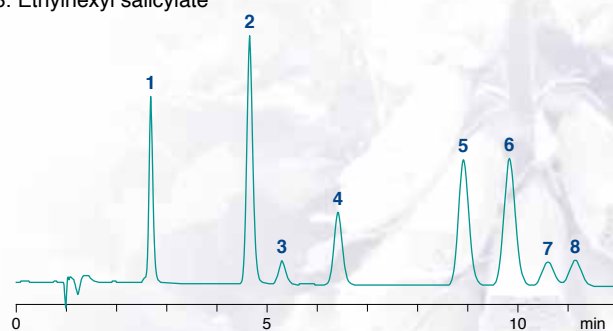
## Sunscreen ingredients

**MN Appl. No. 121500**

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
 Eluent: methanol – 0.5% H<sub>3</sub>PO<sub>4</sub> (82:18, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 42 °C  
 Detection: UV, 300 nm  
 Injection volume: 10 µL

**Peaks:**

1. Benzimidazolecarboxylic acid
2. Benzophenone-3
3. 4-Methylbenzylidene camphor
4. Octocrylene
5. Ethylhexyldimethyl PABA
6. Ethylhexyl methoxycinnamate
7. Butyl methoxydibenzoylmethane (BMDBM)
8. Ethylhexyl salicylate



For separation on C<sub>18</sub> Gravity see appl. 122660 at [www.mn-net.com](http://www.mn-net.com).

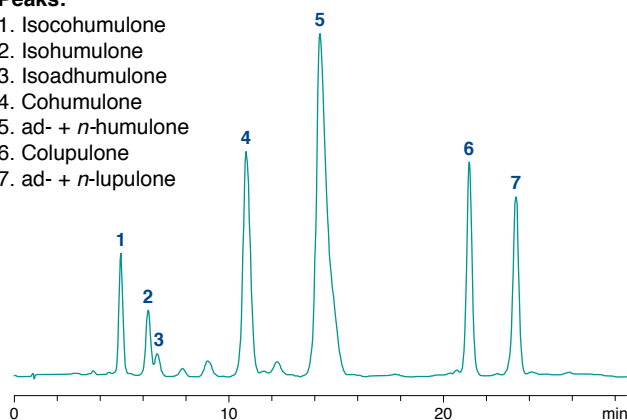
## Determination of iso-alpha-acids, alpha- and beta-acids in isomerized hop pellets

**MN Appl. No. 121100**

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
 Eluent: A) methanol, B) methanol – water – H<sub>3</sub>PO<sub>4</sub> (75:24:1, v/v/v); 100% B (17 min) → 65% B in 8 min → 100% B in 5 min  
 Flow rate: 1.0 mL/min  
 Temperature: 35 °C  
 Detection: UV, 9 min 270 nm, then 314 nm

**Peaks:**

1. Isocohumulone
2. Isohumulone
3. Isoadhumulone
4. Cohumulone
5. ad- + n-humulone
6. Colupulone
7. ad- + n-lupulone



M. Biendl et al., European Brewery Convention, J. of the Institute of Brewing **110** (2004) 242–243.

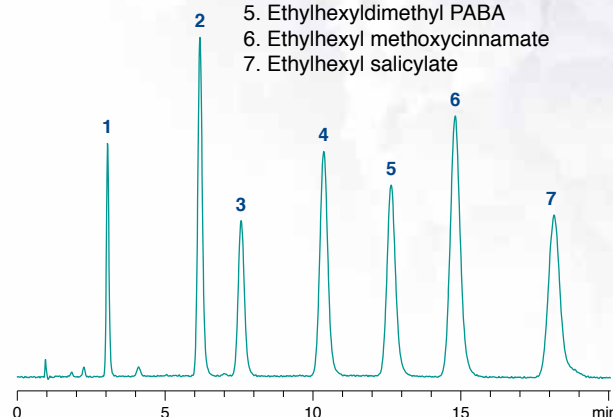
## Sunscreen ingredients

**MN Appl. No. 123640**

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> HTec, 5 µm  
 Eluent: methanol – 100 mM ammonium acetate, pH 4.5 (80:20, v/v)  
 Flow rate: 0.9 mL/min  
 Temperature: 35 °C  
 Detection: UV, 275 nm  
 Injection volume: 12 µL

**Peaks:**

1. Benzophenone
2. 4-Methylbenzylidene camphor
3. Uvinul Plus
4. Octocrylene
5. Ethylhexyldimethyl PABA
6. Ethylhexyl methoxycinnamate
7. Ethylhexyl salicylate



# Applications

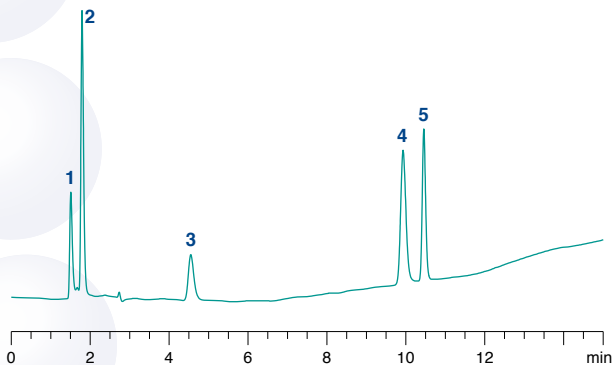
## Water-soluble vitamins

MN Appl. No. 124570

Column: 150 x 3 mm NUCLEODUR® PolarTec, 5 µm  
Eluent: A) 25 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3.0, B) acetonitrile  
10% B (3 min) → 40% B in 12 min  
Flow rate: 0.7 mL/min  
Temperature: 30 °C  
Detection: UV, 220 nm  
Injection volume: 5 µL

### Peaks:

1. Vitamin B<sub>1</sub>
2. Vitamin B<sub>6</sub>
3. Panthotenic acid
4. *p*-Aminobenzoic acid
5. Vitamin B<sub>2</sub>



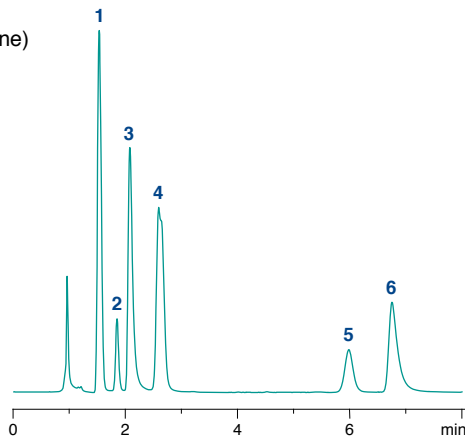
## Water-soluble vitamins

MN Appl. No. 122970

Column: 125 x 4 mm NUCLEODUR® HILIC, 3 µm  
Eluent: A) acetonitrile, B) 25 mM ammonium acetate, pH 4  
80% A (1 min) → 70% A in 11 min  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 30 µL

### Peaks:

1. Nicotinamide
2. Vitamin B<sub>7</sub> (vitamin B<sub>8</sub>, vitamin H, biotin)
3. Vitamin B<sub>6</sub> (pyridoxine)
4. Vitamin C (ascorbic acid)
5. Vitamin B<sub>12</sub> (cyanocobalamin)
6. Vitamin B<sub>1</sub> (thiamine)



## Water-soluble vitamins

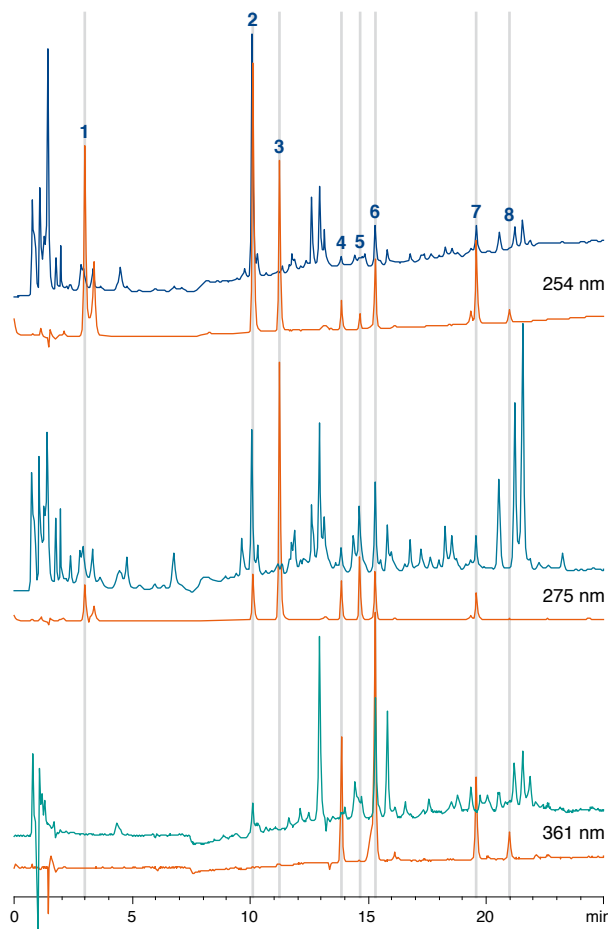
MN Appl. No. 119770

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
Eluent: A) water, 15 mM heptanesulfonic acid (Na salt), 25 mM NaH<sub>2</sub>PO<sub>4</sub>, 0.25% CH<sub>3</sub>COOH, 0.005% triethylamine (pH 3.5), B) acetonitrile – water (40:60, v/v), 15 mM heptanesulfonic acid (Na salt), 0.25% CH<sub>3</sub>COOH, 0.005% triethylamine (pH ~ 3.5); multistep gradient: 0% B (5 min) → 10% B in 2.5 min → 25% B in 2.5 min → 50% B in 8 min → 70% B in 7 min → 0% B in 1 min  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 254, 275 and 361 nm  
Injection volume: 10 µL

### Peaks:

1. Nicotinic acid (0.12 mg/mL)
2. Nicotinamide (0.12 mg/mL)
3. 4-Aminobenzoic acid (0.03 mg/mL)
4. Folic acid (0.24 mg/mL)
5. Vitamin B<sub>6</sub> (pyridoxine hydrochloride, 0.06 mg/mL)
6. Vitamin B<sub>2</sub> (riboflavin, 0.012 mg/mL)
7. Vitamin B<sub>1</sub> (thiamine hydrochloride, 0.06 mg/mL)
8. Rutin (0.012 mg/mL)

orange curves: vitamin test mixture (in eluent A)  
blue curves: multivitamin juice (undiluted)  
both detected at three different wave lengths



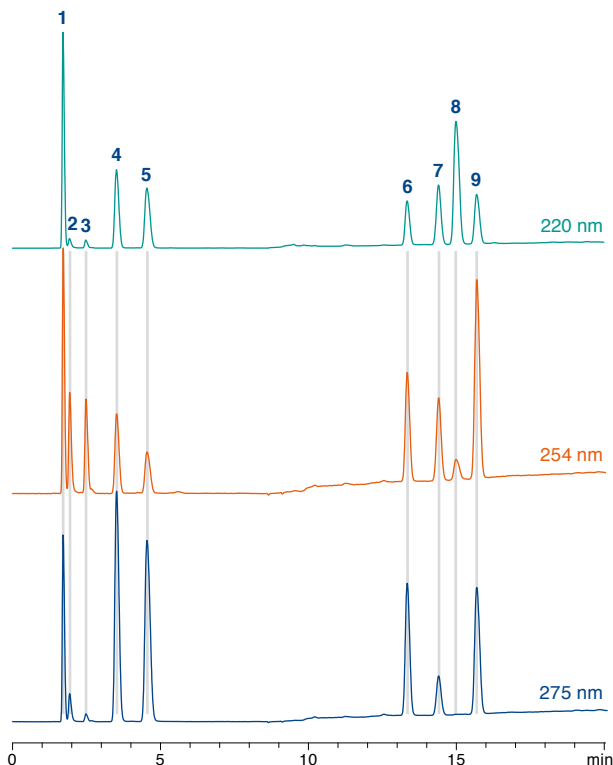
## Water-soluble vitamins

**MN Appl. No. 122450**

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
 Eluent: A) 50 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3,  
 B) methanol – acetonitrile (70:30, v/v)  
 0% B (6 min) → 15% B in 2 min → 35% B in  
 10 min (5 min)  
 Flow rate: 0.6 mL/min  
 Temperature: 40 °C  
 Detection: UV, 218, 254 and 275 nm  
 Injection volume: 10 µL

### Peaks:

1. Vitamin B<sub>1</sub> (thiamine)
2. Pyridoxamine
3. Vitamin C (ascorbic acid)
4. Pyridoxal
5. Vitamin B<sub>6</sub> (pyridoxine)
6. Vitamin B<sub>9</sub> (viamin M, folic acid)
7. Vitamin B<sub>12</sub> (cyanocobalamin)
8. Vitamin B<sub>7</sub> (vitamin B<sub>8</sub>, vitamin H, (+)-biotin)
9. Vitamin B<sub>2</sub> (riboflavin)



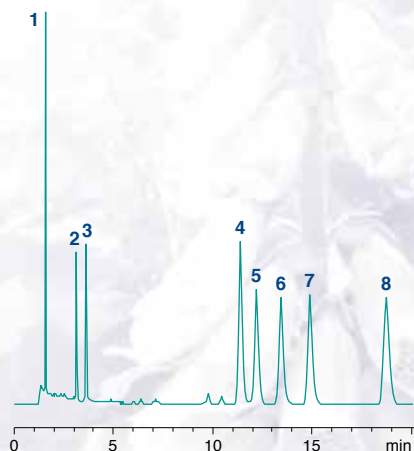
## Fat-soluble vitamins and tocopherols

**MN Appl. No. 117890**

Column: 250 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
 Eluent: acetonitrile  
 Flow rate: 1.5 mL/min  
 Temperature: 30 °C  
 Detection: UV, 280 nm  
 Injection volume: 4 µL

### Peaks:

1. Vitamin K<sub>3</sub>
2. Vitamin A
3. Vitamin A acetate
4. Vitamin D<sub>2</sub>
5. Vitamin D<sub>3</sub>
6. Vitamin E  
(α-tocopherol)
7. Vitamin E acetate  
(α-tocopherol  
acetate)
8. Vitamin K<sub>1</sub>

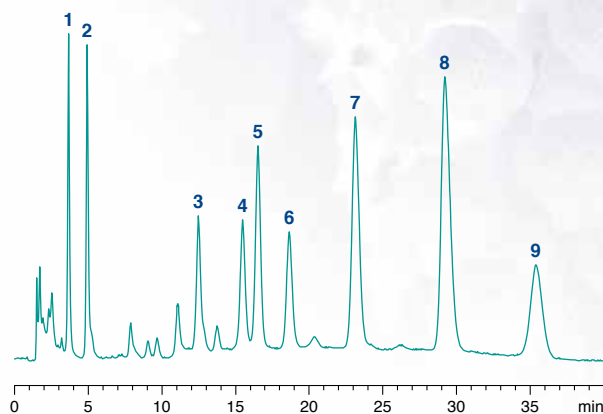


**MN Appl. No. 121160**

Column: 125 x 2 mm NUCLEODUR® C<sub>18</sub> Isis, 5 µm  
 Eluent: acetonitrile – water (100:5, v/v)  
 Flow rate: 0.2 mL/min  
 Temperature: 25 °C  
 Detection: UV, 275 nm  
 Injection volume: 5 µL

### Peaks:

- |                           |   |
|---------------------------|---|
| 1. Vitamin A              | 6. γ-Tocopherol                             |
| 2. Vitamin A acetate      | 7. Vitamin E (α-tocopherol)                 |
| 3. Vitamin K <sub>2</sub> | 8. Vitamin E acetate (α-tocopherol acetate) |
| 4. Vitamin D <sub>2</sub> | 9. Vitamin K <sub>1</sub>                   |
| 5. Vitamin D <sub>3</sub> |   |



For separation of tocopherols on NUCLEODUR® 100-5 C<sub>18</sub> ec see appl. 117910 at [www.mn-net.com](http://www.mn-net.com).

# Applications

## Complexing agents acc. to DIN 38 413-8

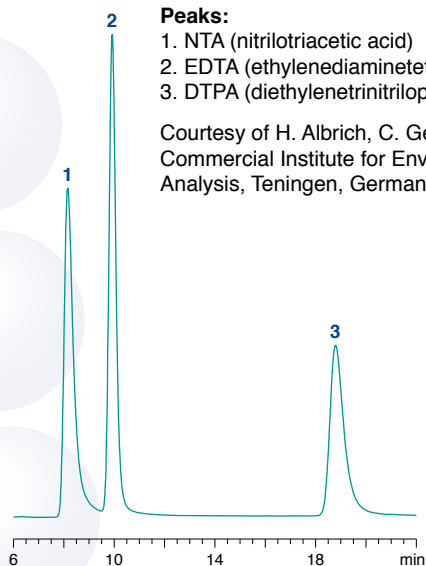
### MN Appl. No. 119780

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
Eluent: 0.6 mM HNO<sub>3</sub>, 7.53 mM N(C<sub>4</sub>H<sub>9</sub>)<sub>4</sub>HSO<sub>4</sub>,  
2.6 mM N(C<sub>4</sub>H<sub>9</sub>)<sub>4</sub>OH, 37 µM Fe<sup>3+</sup>  
Flow rate: 0.6 mL/min  
Temperature: 20 °C  
Detection: UV, 260 nm  
Injection volume: 50 µL

#### Peaks:

1. NTA (nitrilotriacetic acid)
2. EDTA (ethylenediaminetetraacetic acid)
3. DTPA (diethylenetrinitriolpentaacetic acid)

Courtesy of H. Albrich, C. Geis, GIU;  
Commercial Institute for Environmental  
Analysis, Teningen, Germany.



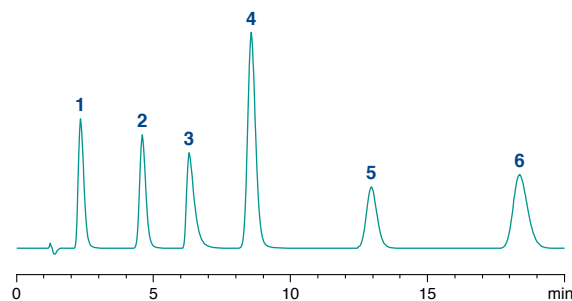
## Aromatic acids

### MN Appl. No. 121180

Column: 125 x 2 mm NUCLEODUR® C<sub>18</sub> Isis, 5 µm  
Eluent: methanol – 50 mM KH<sub>2</sub>PO<sub>4</sub>, pH 3 (10:90, v/v)  
Flow rate: 0.25 mL/min  
Temperature: 30 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

#### Peaks:

1. Gallic acid
2. 3,4-Dihydroxybenzoic acid
3. 2,5-Dihydroxybenzoic acid
4. 4-Hydroxybenzoic acid
5. Syringic acid
6. Vanillic acid



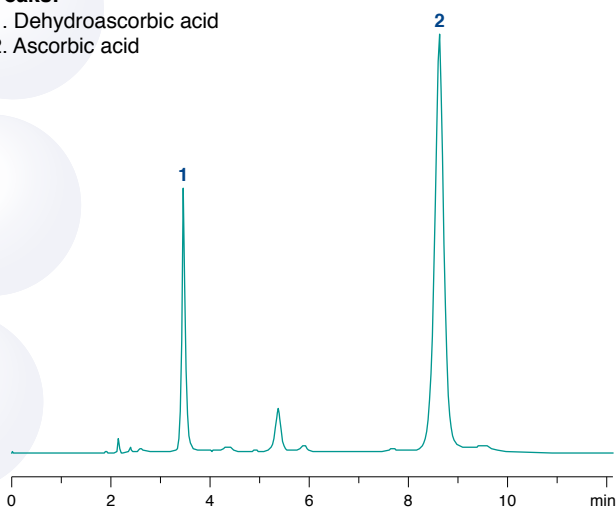
## Ascorbic acid and dehydroascorbic acid

### MN Appl. No. 122940

Column: 250 x 4 mm NUCLEODUR® HILIC, 5 µm  
Eluent: acetonitrile – 100 mM ammonium acetate  
(70:30, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 240 nm

#### Peaks:

1. Dehydroascorbic acid
2. Ascorbic acid



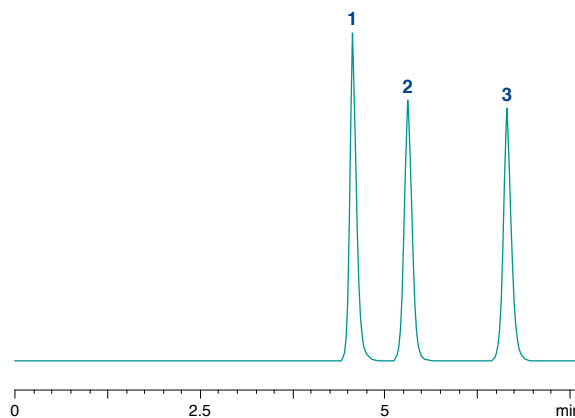
## Organic acids

### MN Appl. No. 119290

Column: 250 x 4 mm NUCLEODUR® 100-5 CN-RP  
Eluent: 25 mM KH<sub>2</sub>PO<sub>4</sub>, pH 4.0  
Flow rate: 0.5 mL/min  
Temperature: 30 °C  
Detection: UV, 210 nm  
Injection volume: 15 µL

#### Peaks:

1. Aspartic acid
2. Fumaric acid
3. Maleic acid





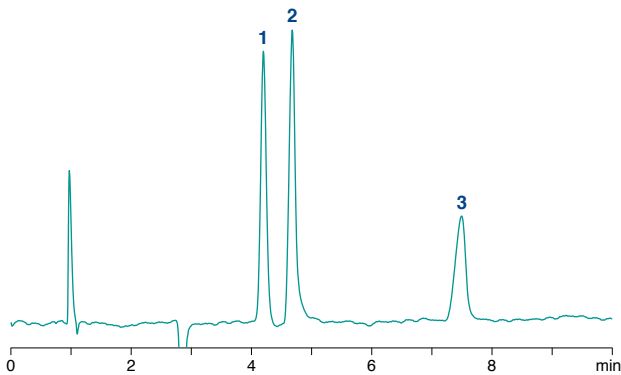
## Organic acids

**MN Appl. No. 122930**

Column: 125 x 4 mm NUCLEODUR® HILIC, 3 µm  
 Eluent: acetonitrile – 200 mM ammonium acetate, pH 6.8 (70:30, v/v)  
 Flow rate: 1 mL/min  
 Temperature: 25 °C  
 Detection: UV, 220 nm  
 Injection volume: 0.5 µL

### Peaks:

1. Fumaric acid
2. Oxalic acid
3. Citric acid

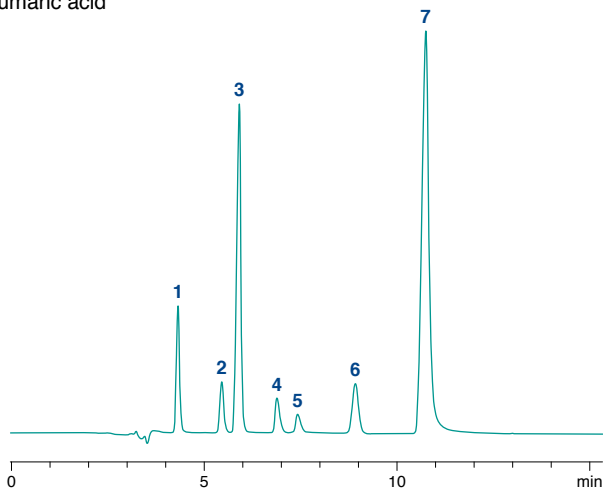


**MN Appl. No. 120500**

Column: 250 x 4.6 mm NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm  
 Eluent: 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 2.6  
 Flow rate: 0.7 mL/min  
 Detection: UV, 210 nm  
 Injection volume: 20 µL

### Peaks:

1. Tartaric acid
2. Malic acid
3. Shikimic acid
4. Lactic acid
5. Acetic acid
6. Citric acid
7. Fumaric acid



Also see appl. 119180 at [www.mn-net.com](http://www.mn-net.com).

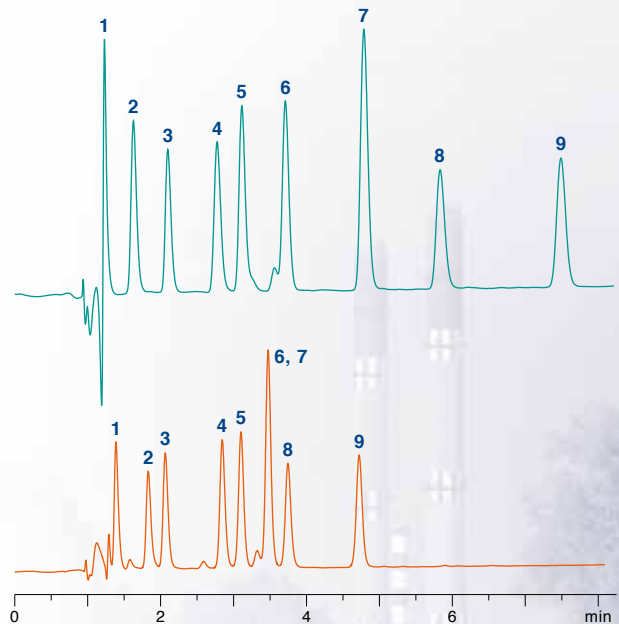
## Organic acids

**MN Appl. No. 124562**

Columns: 150 x 3 mm NUCLEODUR® PolarTec, 5 µm  
 150 x 3 mm Waters SymmetryShield™ RP18, 5 µm  
 Eluent: A) acetonitrile, 0.1 % TFA, B) water, 0.1 % TFA  
 20 % A → 60 % A in 12 min  
 Flow rate: 0.73 mL/min  
 Temperature: 20 °C  
 Detection: UV, 254 nm  
 Injection volume: 5 µL

### Peaks:

- |                               |                              |
|-------------------------------|------------------------------|
| 1. Dihydroxymandelic acid     | 6. Vanillic acid             |
| 2. Gallic acid                | 7. 3-Hydroxybenzoic acid     |
| 3. Dihydroxyphenylacetic acid | 8. 2,5-Dihydroxybenzoic acid |
| 4. 3,4-Dihydroxybenzoic acid  | 9. 2,4-Dihydroxybenzoic acid |
| 5. Syringic acid              |                              |



# Applications

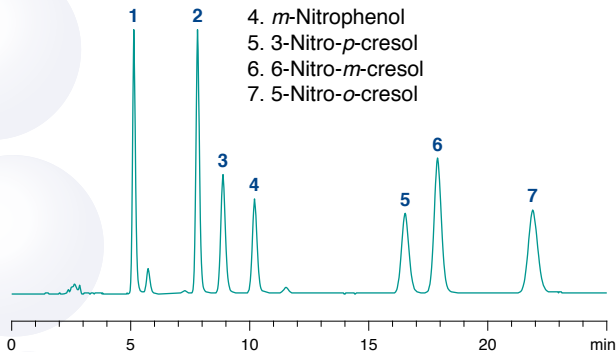
## Nitrophenols

MN Appl. No. 122650

Column: 250 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
 Eluent: methanol – 20 mM NaH<sub>2</sub>PO<sub>4</sub>, pH 5 (49:51, v/v)  
 Flow rate: 0.8 mL/min  
 Temperature: 20 °C  
 Detection: UV, 235 nm  
 Injection volume: 2.0 µL

### Peaks:

1. *p*-Nitrophenol
2. *o*-Nitrophenol
3. 4-Nitro-*m*-cresol
4. *m*-Nitrophenol
5. 3-Nitro-*p*-cresol
6. 6-Nitro-*m*-cresol
7. 5-Nitro-*o*-cresol



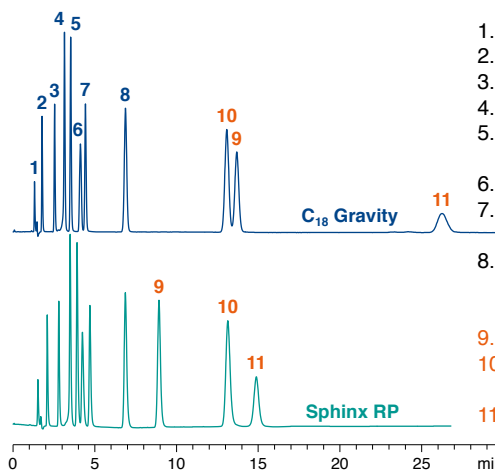
## Substituted aromatics

MN Appl. No. 119840/119850

Columns: 150 x 4.6 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
 150 x 4.6 mm NUCLEODUR® Sphinx RP, 5 µm  
 Eluent: methanol – water (55:45, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm  
 Injection volume: 2 µL

### Peaks:

1. Uracil
2. Benzamide
3. Phenol
4. Benzaldehyde
5. Acetophenone
6. 2-Nitrophenol
7. Nitrobenzene
8. Propyl 4-hydroxybenzoate
9. Toluene
10. Benzophenone
11. Xylene



Selectivity comparison of NUCLEODUR® C<sub>18</sub> Gravity and Sphinx RP shows lower hydrophobicity and retention of Sphinx RP for substituted aromatics like toluene and xylene.

## Nitroaromatics EPA 8330

MN Appl. No. 124490/124500

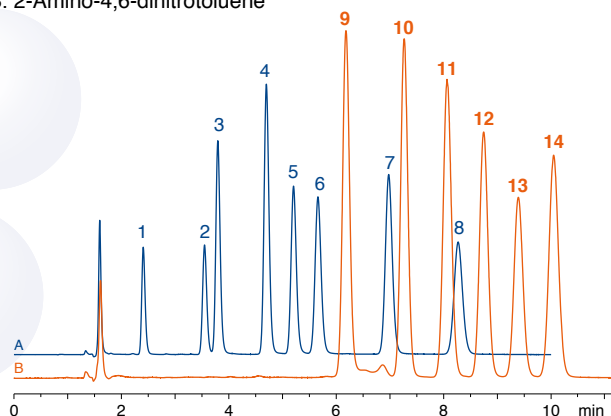
Column: 150 x 3 mm NUCLEODUR® PolarTec, 5 µm  
 Eluent: Mix A: water – methanol (50:50, v/v)  
 Mix B: water, 0.1 % formic acid – methanol (45:55, v/v)  
 Flow rate: 0.46 mL/min  
 Temperature: 50 °C, 60 °C  
 Detection: UV, 254 nm  
 Injection volume: 5 µL

### Peaks Mix A:

1. Octogen (HMX)
2. Hexogen (RDX)
3. 1,3,5-Trinitrobenzene
4. 1,3-Dinitrobenzene
5. Nitrobenzene
6. 2,4,6-Trinitrotoluene
7. 2,4-Dinitrotoluene
8. 2-Amino-4,6-dinitrotoluene

### Peaks Mix B:

9. *N*-Methyl-*N*-2,4,6-tetranitroaniline (Tetryl)
10. 4-Amino-2,6-dinitrotoluene
11. 2,6-Dinitrotoluene
12. 2-Nitrotoluene
13. 4-Nitrotoluene
14. 3-Nitrotoluene



For separation of Mix A and Mix B in a single run see appl. 124510 at [www.mn-net.com](http://www.mn-net.com).

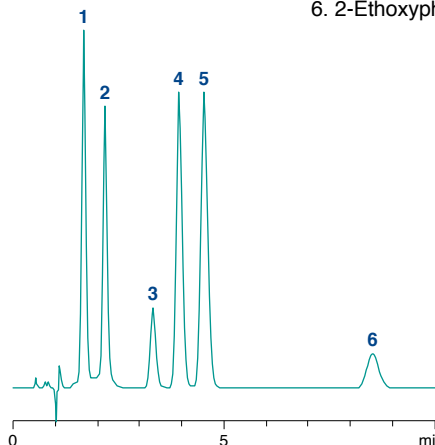
## Phenolic compounds

MN Appl. No. 117970

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
 Eluent: methanol – water, 0.1 % H<sub>3</sub>PO<sub>4</sub> (40:60, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 22 °C  
 Detection: UV, 254 nm  
 Injection volume: 5 µL

### Peaks:

1. Resorcinol
2. Pyrocatechol
3. 4-Methoxyphenol
4. Phenol
5. 2-Methoxyphenol
6. 2-Ethoxyphenol



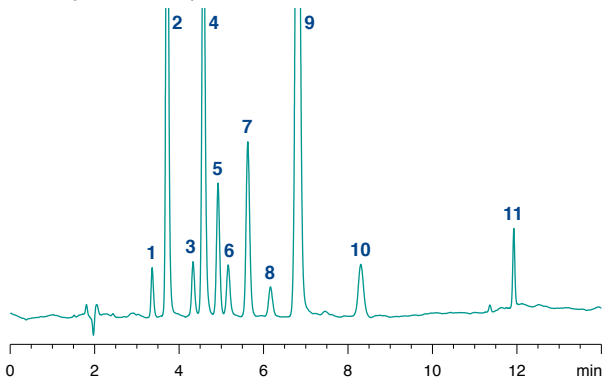
## Phenolic compounds

MN Appl. No. 124740

Column: 250 x 3 mm NUCLEODUR® PFP, 5 µm  
 Eluent: A) acetonitrile + 1 % acetic acid  
 B) water + 1 % acetic acid  
 Gradient: 45 % A (7 min) → 70 % A in 2 min (5 min)  
 Flow rate: 0.6 mL/min  
 Temperature: 45 °C  
 Detection: UV, 254 nm  
 Injection volume: 1 µL

### Peaks:

- |                               |                            |
|-------------------------------|----------------------------|
| 1. Phenol                     | 7. 4-Chloro-3-methylphenol |
| 2. 4-Nitrophenol              | 8. 2,4-Dichlorophenol      |
| 3. 2-Nitrophenol              | 9. 2,4-Dimethylphenol      |
| 4. 2,4-Dinitrophenol          | 10. 2,4,6-Trichlorophenol  |
| 5. 2-Chlorophenol             | 11. Pentachlorophenol      |
| 6. 2-Methyl-4,6-dinitrophenol |                            |



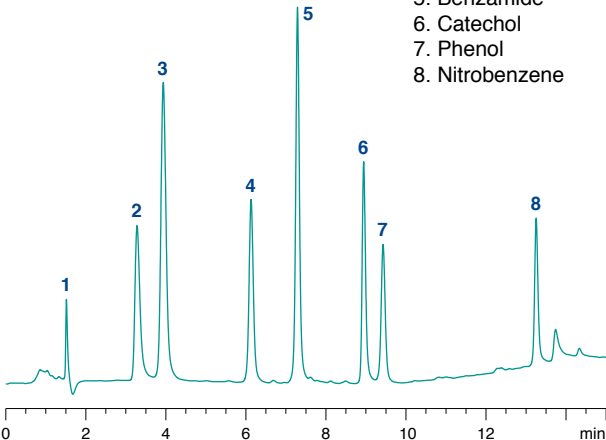
## Phenolic compounds

MN Appl. No. 124750

Column: 100 x 2 mm NUCLEODUR® PFP, 3 µm  
 Eluent: A) methanol, B) 10 mM ammonium acetate, pH 6.8  
 5 % A → 80 % A in 15 min  
 Flow rate: 0.25 mL/min  
 Temperature: 18 °C  
 Detection: UV, 230 nm  
 Injection volume: 1 µL

### Peaks:

1. Uracil
2. Pyrogallol
3. Phloroglucinol
4. Resorcinol
5. Benzamide
6. Catechol
7. Phenol
8. Nitrobenzene



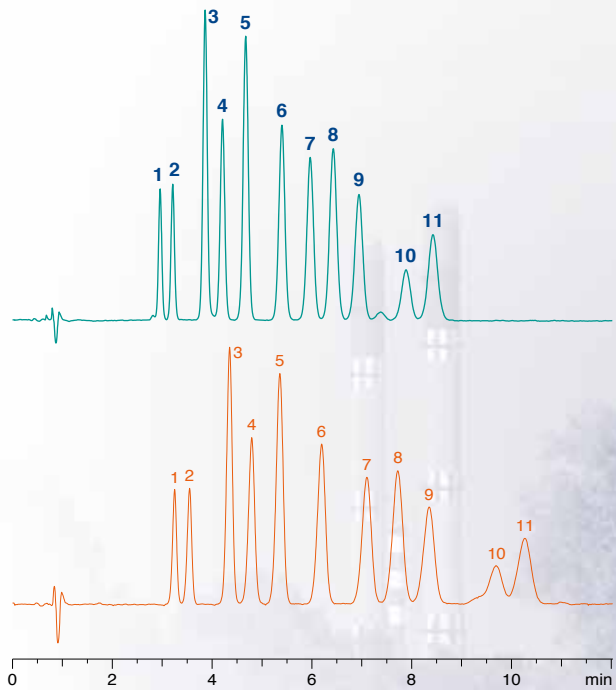
## Separation of phenol isomers

MN Appl. No. 124541

Columns: 100 x 4.6 mm NUCLEODUR® PFP, 5 µm  
 100 x 4.6 mm Phenomenex Luna® PFP(2), 5 µm  
 Eluent: acetonitrile, 0.1 % formic acid – water, 0.1 %  
 formic acid (35:65, v/v)  
 Flow rate: 1.3 mL/min  
 Temperature: 35 °C  
 Detection: UV, 280 nm

### Peaks:

- |                       |                       |
|-----------------------|-----------------------|
| 1. <i>o</i> -Cresol   | 7. 2,3-Dichlorophenol |
| 2. <i>m</i> -Cresol   | 8. 2,4-Dichlorophenol |
| 3. 3,4-Dimethylphenol | 9. 3,4-Dichlorophenol |
| 4. 3,5-Dimethylphenol | 10. 2,4-Dibromophenol |
| 5. 2,5-Dimethylphenol | 11. 3,5-Dibromophenol |
| 6. 2,6-Dichlorophenol |                       |



NUCLEODUR® PFP provides under identical conditions a better separation than Luna® PFP(2). While on Luna® PFP(2) for peaks 10 and 11 only a resolution of 1.27 is obtained, on NUCLEODUR® PFP the peaks are baseline separated ( $R_S = 1.56$ ).

# Applications

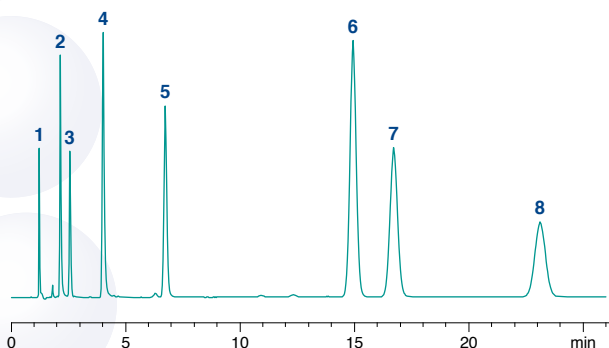
## Amines

### MN Appl. No. 121200

Column: 150 x 4.6 mm NUCLEODUR® C<sub>18</sub> Isis, 5 µm  
Eluent: acetonitrile – 50 mM K<sub>2</sub>HPO<sub>4</sub> (40:60, v/v), pH 8  
Flow rate: 1 mL/min  
Temperature: 25 °C  
Detection: UV, 254 nm  
Injection volume: 8 µL

#### Peaks:

1. Uracil
2. Pyridine
3. Desethylatrazine
4. 4-Acetylpyridine
5. 4-Ethylaniline
6. *N,N*-Dimethylaniline
7. 4-Aminoanthraquinone
8. 3,5-Dinitro-(1-phenylethyl)benzamide



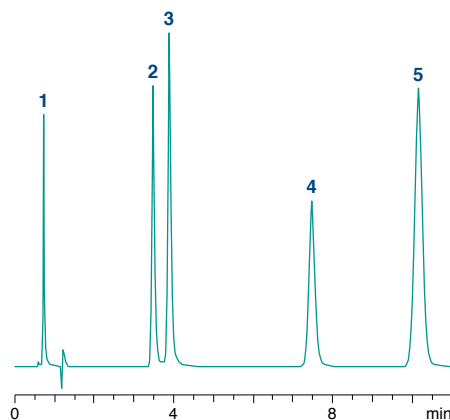
## Aromatic aldehydes

### MN Appl. No. 117990

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
Eluent: acetonitrile – water, pH 6.0 (22:78, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 22 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL (~10–50 µg/mL)

#### Peaks:

1. *p*-Carboxybenzaldehyde
2. *p*-Hydroxybenzaldehyde
3. Vanillin
4. 4-Ethoxyvanillin
5. Benzaldehyde



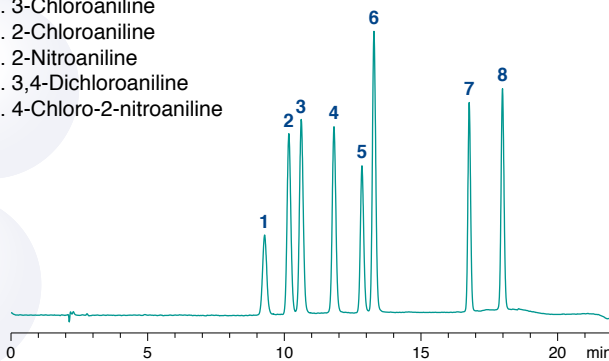
## Aromatic amines

### MN Appl. No. 124800

Column: 250 x 4 mm NUCLEODUR® PFP, 5 µm  
Eluent: A) methanol, B) 20 mM K<sub>2</sub>HPO<sub>4</sub>, pH 3  
40% A (5 min) → 70% A in 10 min (7 min)  
Flow rate: 1 mL/min  
Temperature: 20 °C  
Detection: UV, 254 nm  
Injection volume: 1 µL

#### Peaks:

1. 4-Chloroaniline
2. 4-Nitroaniline
3. 3-Nitroaniline
4. 3-Chloroaniline
5. 2-Chloroaniline
6. 2-Nitroaniline
7. 3,4-Dichloroaniline
8. 4-Chloro-2-nitroaniline



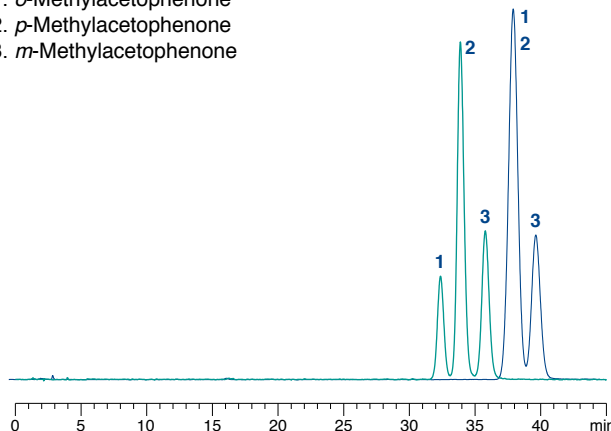
## Aromatic ketones

### MN Appl. No. 124761

Columns: 250 x 4 mm NUCLEODUR® PFP, 5 µm  
250 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Eluent: methanol – water (35:65, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 35 °C  
Detection: UV, 254 nm  
Injection volume: 1 µL

#### Peaks:

1. *o*-Methylacetophenone
2. *p*-Methylacetophenone
3. *m*-Methylacetophenone



Distinct steric selectivity of NUCLEODUR® PFP provides separation of all three regioisomers, while on NUCLEODUR® C<sub>18</sub> Gravity baseline separation can't be achieved.

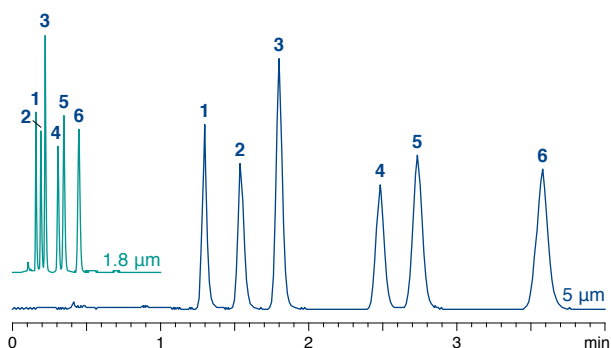
## Aromatic ketones

**MN Appl. No. 122720/122730**

Column: 125 x 2 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
 50 x 2 mm NUCLEODUR® C<sub>18</sub> Gravity, 1.8 µm  
 Eluent: acetonitrile – water (60:40, v/v)  
 Flow rate: 0.33 mL/min, 1.25 mL/min  
 Temperature: 25 °C  
 Detection: UV, 230 nm

### Peaks:

1. Acetophenone
2. Eugenol
3. Propiophenone
4. Butyrophenone
5. Benzophenone
6. Valerophenone

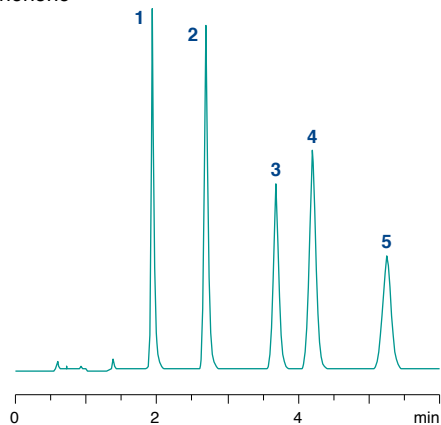


**MN Appl. No. 117980**

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
 Eluent: acetonitrile – water (60:40, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 22 °C  
 Detection: UV, 230 nm  
 Injection volume: 2 µL (~10–50 µg/mL)

### Peaks:

1. Acetophenone
2. Propiophenone
3. Butyrophenone
4. Benzophenone
5. Valerophenone



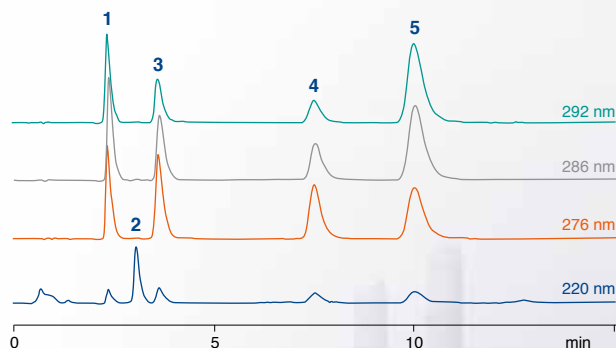
## Furfurol and related compounds in transformer oil in accordance with DIN EN 61198-B

**MN Appl. No. 121662**

Column: 125 x 4 mm NUCLEODUR® 100-5 C<sub>18</sub> ec  
 Sample prep.: SPE see appl. 304180 at [www.mn-net.com](http://www.mn-net.com)  
 Eluent: methanol – water (10:90, v/v)  
 Flow rate: 2.0 mL/min  
 Detection: UV, 220, 276, 286 and 292 nm

### Peaks:

1. 5-Hydroxymethyl-2-furfurol
2. 2-Furfuryl alcohol
3. 2-Furfurol
4. 2-Acetylfuran
5. 5-Methyl-2-furfurol



A. Heiseler et al., GIT (2004) 504–505

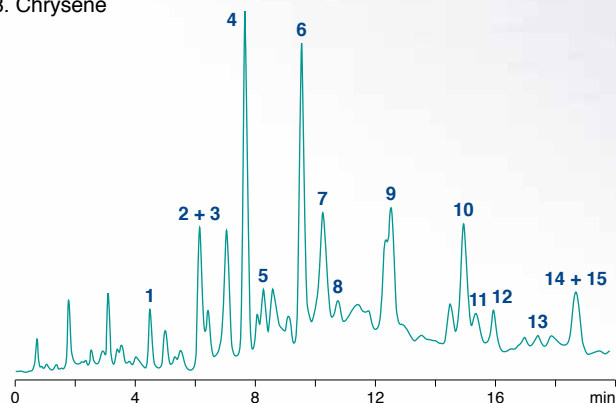
## PAHs from tar

**MN Appl. No. 120740**

Column: 150 x 4.6 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
 Sample prep.: see Appl. 120740 at [www.mn-net.com](http://www.mn-net.com)  
 Eluent: A) water, B) acetonitrile  
 60% B (3 min) → 100% B in 27 min  
 Flow rate: 1.0 mL/min  
 Detection: UV, 220 nm

### Peaks:

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



# Applications

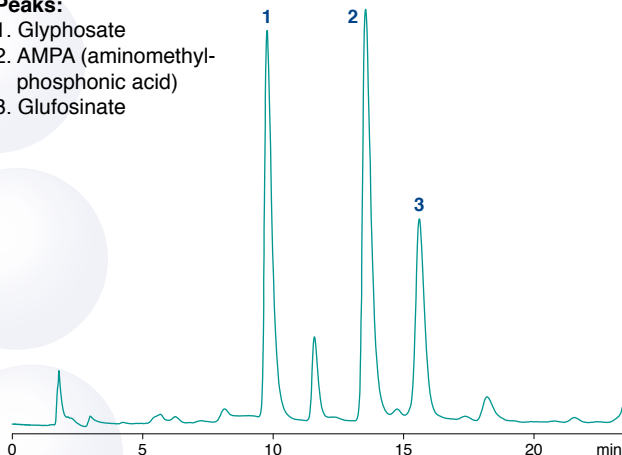
## Organophosphorus herbicides

### MN Appl. No. 120490

Column: 250 x 3 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
Sample prep.: for SPE see Appl. 303780 at [www.mn-net.com](http://www.mn-net.com);  
derivatization with FMOC-Cl  
Eluent: A) acetonitrile, B) H<sub>3</sub>PO<sub>4</sub>, pH 1.2  
30% A → 35% A in 27 min → 90% A in 3 min  
(6 min) → 30% A in 2 min (7 min)  
Flow rate: 0.5 mL/min  
Temperature: 30 °C  
Detection: fluorescence, λ<sub>ex</sub> 263 nm, λ<sub>em</sub> 317 nm

#### Peaks:

1. Glyphosate
2. AMPA (aminomethylphosphonic acid)
3. Glufosinate



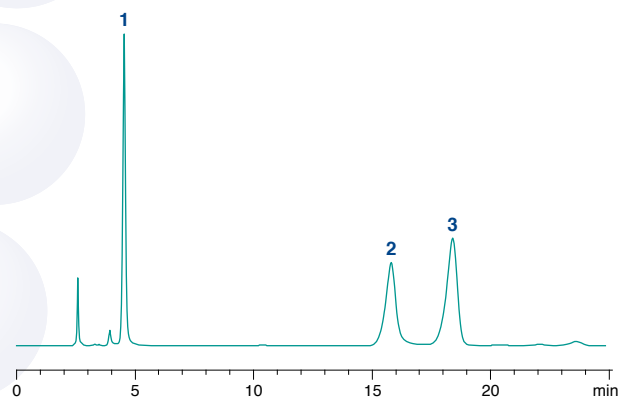
Courtesy of Mr. Schüssler, Mrs. Mikler, Bavarian State Agency for Water Management, Munich.

### MN Appl. No. 122190

Column: 250 x 4 mm NUCLEODUR® 100-5 NH<sub>2</sub>-RP  
Sample prep.: derivatization with FMOC, concentration of each pesticide 0.3 mg/mL  
Eluent: acetonitrile – 50 mM KH<sub>2</sub>PO<sub>4</sub>, pH 4.6 (60:40, v/v)  
Flow rate: 0.8 mL/min  
Temperature: 40 °C  
Detection: UV, 254 nm  
Injection volume: 5 µL

#### Peaks:

1. AMPA
2. Glyphosate
3. Glufosinate



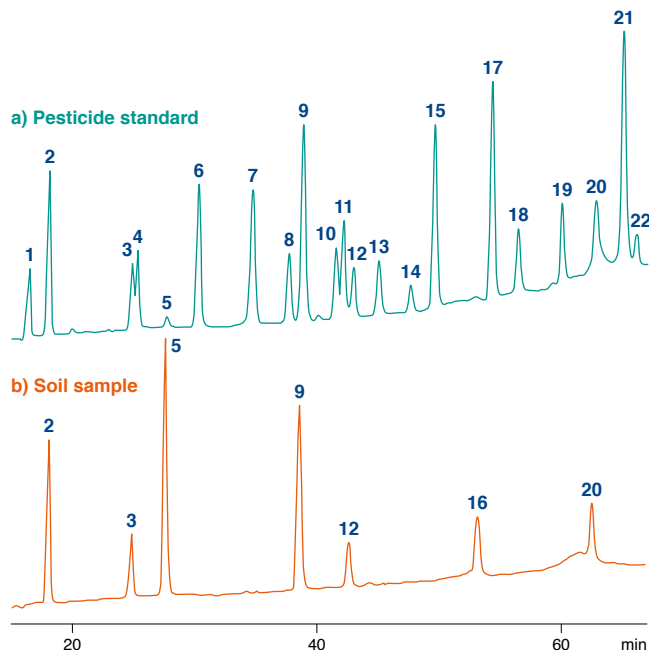
## Pesticides from soil

### MN Appl. No. 119890

Column: 250 x 4 mm NUCLEODUR® 100-3 C<sub>8</sub> ec  
Eluent: A) water, B) acetonitrile,  
10% B → 25% B in 10 min → 30% B in  
10 min (5 min) → 40% B in 20 min → 50% B in  
20 min (10 min)  
Flow rate: 0.8 mL/min  
Temperature: 35 °C  
Detection: UV, 230 nm

#### Peaks:

1. Metamitron
2. Desethylatrazine
3. Hexazinone
4. Metoxuron
5. Simazine
6. Cyanazine
7. Methabenzthiazuron (Tribunil®)
8. Chlortoluron
9. Atrazine
10. Monolinuron
11. Isoproturon
12. Diuron
13. Metobromuron
14. Metazachlor
15. Sebuthylazine
16. Dichlobenil
17. Terbutylazine
18. Linuron
19. Chloroxuron
20. Propyzamid
21. Terbutryn
22. Metolachlor



Courtesy of E. Marek, LUFA Center for Analyses, Münster, Germany.

Also see appl. 118010 at [www.mn-net.com](http://www.mn-net.com).

# Pollutants and miscellaneous organics

## Pesticides

MN Appl. No. 120481: triazines

MN Appl. No. 120482: phenylurea derivatives

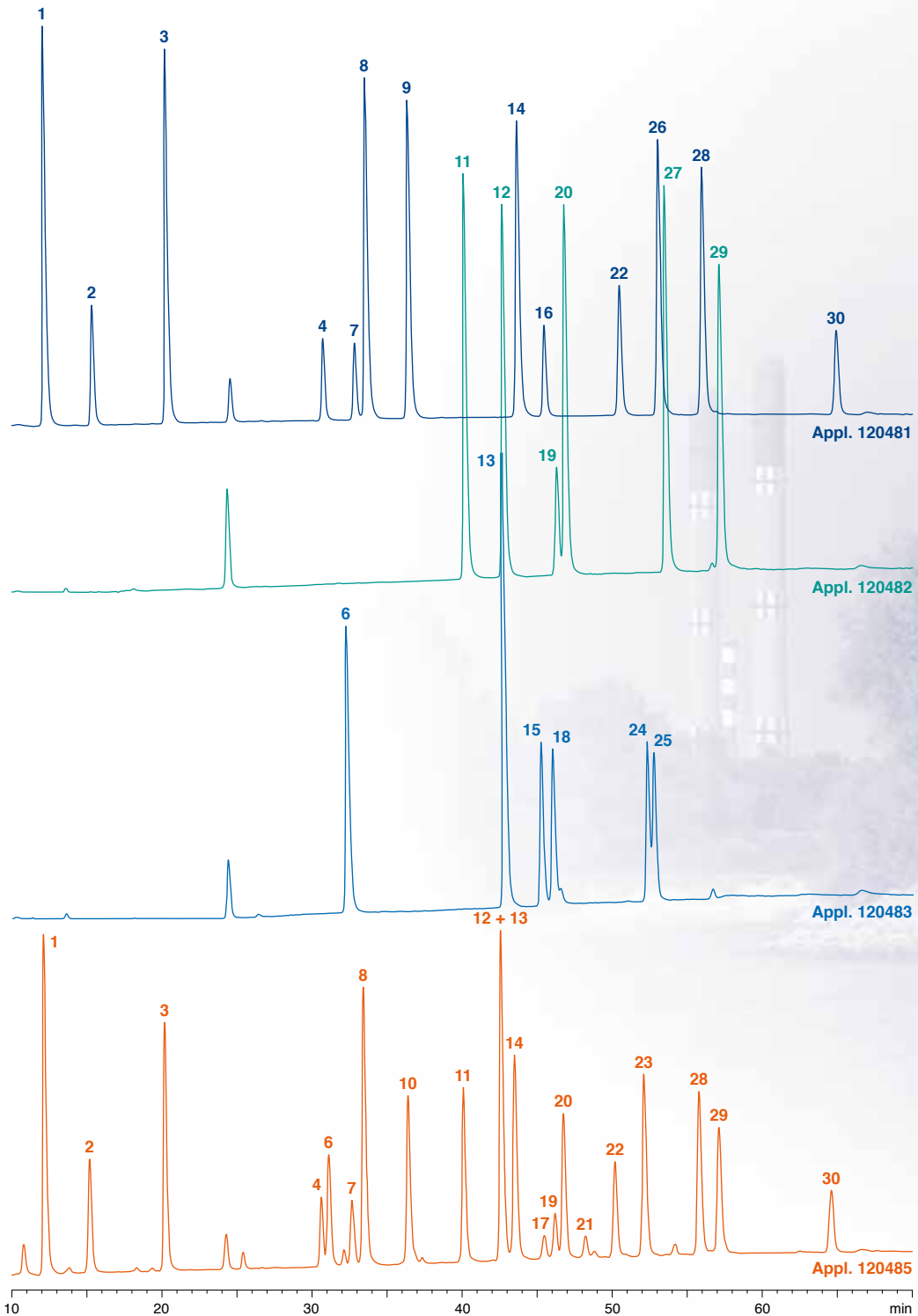
MN Appl. No. 120483: phenoxy-carboxylic acids

MN Appl. No. 120485: 21 pesticides

Column: 250 x 4 mm NUCLEODUR® 100-3 C<sub>8</sub> ec with 8 x 4 mm guard column  
Eluent: A) acetonitrile, B) 20 mM KH<sub>2</sub>PO<sub>4</sub> + 1 mL conc. H<sub>3</sub>PO<sub>4</sub>, 85% B (5 min) → 47% B in 60 min  
Flow rate: 0.7 mL/min  
Temperature: 35 °C  
Detection: UV, 218 nm  
Injection volume: 50 µL

### Peaks:

1. Desisopropylatrazine
2. 2,4-Dichlorobenzamide
3. Desethylatrazine
4. Hexazinone
5. Metoxuron
6. Dicamba
7. Bromacil
8. Simazine
9. Desethylterbutylazine
10. Cyanazine
11. Methabenzthiazuron
12. Chlortoluron
13. Bentazone
14. Atrazine
15. 2,4-D
16. Metalaxyl
17. Monolinuron
18. MCPA
19. Isoproturon
20. Diuron
21. Metobromuron
22. Metazachlor
23. Sebuthylazine
24. Dichlorprop
25. Mecoprop
26. Propazine
27. Dimefuron
28. Terbutylazine
29. Linuron
30. Metolachlor



Courtesy of C. Geis, GIU;  
Commercial Institute for  
Environmental Analysis,  
Teningen, Germany.

# Applications

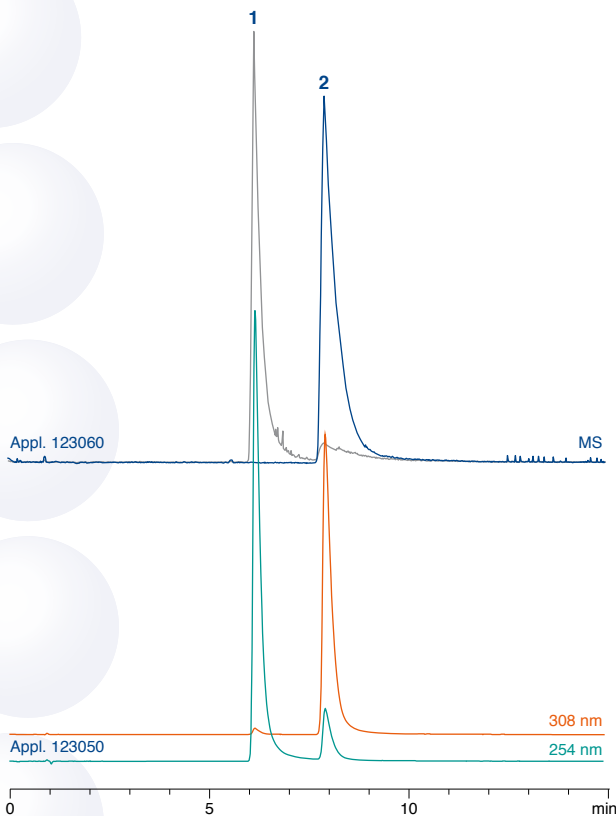
## Herbicides

**MN Appl. No. 123050/123060**

Column: 125 x 2 mm NUCLEODUR® HILIC, 3 µm  
Eluent: acetonitrile – 50 mM ammonium formate, pH 3.2 (80:20, v/v)  
Flow rate: 0.3 mL/min  
Temperature: 45 °C  
Detection: UV, 254 and 308 nm; MS  
Injection volume: 1 µL, 0.5 mg/mL

### Peaks:

1. Paraquat
2. Diquat



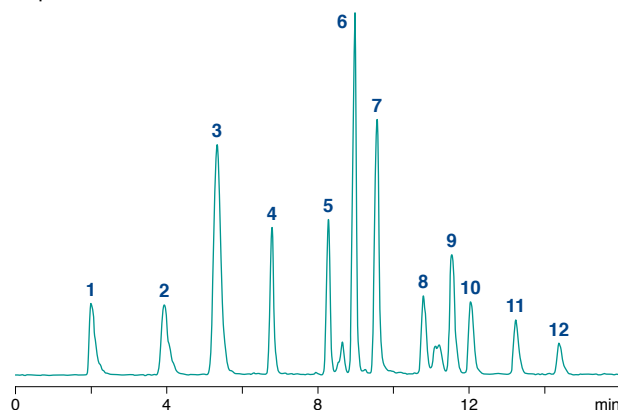
## Perfluorinated surfactants in water

**MN Appl. No. 121590**

Column: 125 x 2 mm NUCLEODUR® Sphinx RP, 3 µm  
Sample prep.: see appl. 121590 at [www.mn-net.com](http://www.mn-net.com)  
Eluent: A) 10 mM NH<sub>4</sub> acetate in water – methanol (75:25, v/v); B) 10 mM NH<sub>4</sub> acetate in acetonitrile – methanol (75:25, v/v); 10% B → 30% B in 3 min → 55% B in 8 min → 70% B in 4 min  
Flow rate: 0.3 mL/min; temperature 50 °C  
Detection: LC-MS-MS; injection volume 50 µL

### Peaks:

- |                               |                               |
|-------------------------------|-------------------------------|
| 1. Perfluorobutanoic acid     | 7. Perfluorooctanoic acid     |
| 2. Perfluoropentanoic acid    | 8. Perfluorononanoic acid     |
| 3. K perfluorobutanesulfonate | 9. K perfluorooctanesulfonate |
| 4. Perfluorohexanoic acid     | 10. Perfluorodecanoic acid    |
| 5. Perfluoroheptanoic acid    | 11. Perfluoroundecanoic acid  |
| 6. K perfluorohexansulfonate  | 12. Perfluorododecanoic acid  |



D. Skutlarek et al., Environ Sci Pollut Res 13 (2006) 299–307

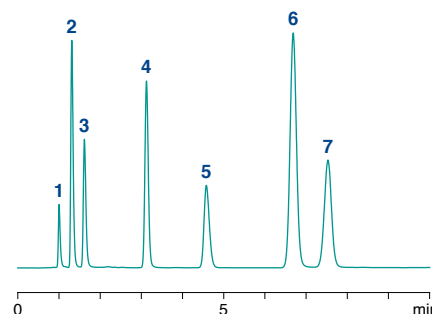
## Selectivity test

**MN Appl. No. 119880**

Column: 125 x 4 mm NUCLEODUR® Sphinx RP, 5 µm  
Eluent: methanol – 25 mM NH<sub>4</sub>H<sub>2</sub>PO<sub>4</sub>, pH 7 (65:35, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 40 °C  
Detection: UV, 254 nm  
Injection volume: 6 µL

### Peaks:

1. Uracil
2. 2,7-Dihydroxynaphthalene
3. 2,3-Dihydroxynaphthalene
4. Ethyl benzoate
5. Lidocaine
6. Biphenyl
7. Acenaphthene





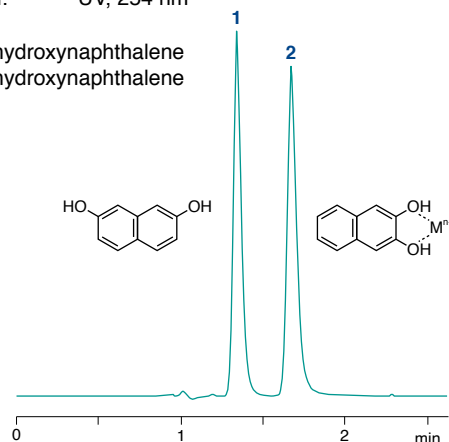
## Test for metal ions in silica adsorbent

**MN Appl. No. 118630**

Column: 125 x 4 mm NUCLEODUR® C<sub>8</sub> Gravity, 5 µm  
 Eluent: methanol – 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 7 (65:35, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 25 °C  
 Detection: UV, 254 nm

**Peaks:**

1. 2,7-Dihydroxynaphthalene
2. 2,3-Dihydroxynaphthalene



The ratio of the asymmetry factors of 2,3-dihydroxynaphthalene (2) and 2,7-dihydroxynaphthalene (1) is a measure for the metal ion content of the silica phase, because (2) can form complexes with metal ions, resulting in broad peaks for this compound.

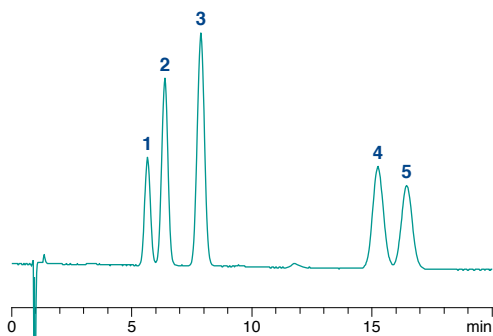
## Dihydroxynaphthalenes

**MN Appl. No. 121190**

Column: 125 x 4 mm NUCLEODUR® C<sub>18</sub> Isis, 5 µm  
 Eluent: methanol – 0.5% H<sub>3</sub>PO<sub>4</sub> (30:70, v/v)  
 Flow rate: 1 mL/min  
 Temperature: 30 °C  
 Detection: UV, 254 nm  
 Injection volume: 15 µL

**Peaks:**

1. 1,5-Dihydroxynaphthalene
2. 1,6-Dihydroxynaphthalene
3. 2,7-Dihydroxynaphthalene
4. 1,3-Dihydroxynaphthalene
5. 2,3-Dihydroxynaphthalene



# Substance index

## A

Acenaphthene	C <sub>18</sub> Gravity, 5 µm	9
	C <sub>18</sub> Pyramid, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	15
	Sphinx RP, 5 µm	62
Acenaphthylene	C <sub>18</sub> Gravity, 5 µm	59
Acesulfame K	100-5 C <sub>8</sub> ec	43
	C <sub>18</sub> HTec, 5 µm	43
2-Acetamidophen	C <sub>8</sub> Gravity, 5 µm	32
Acetamidophenol isomers	C <sub>18</sub> HTec, 5 µm	23
Acetaminophen	see Paracetamol	
Acetanilide	C <sub>8</sub> Gravity, 5 µm	32
Acetic acid	C <sub>18</sub> Pyramid, 5 µm	14, 15, 55
Acetophenone	100-5 C <sub>18</sub> ec	59
	C <sub>18</sub> Gravity, 1.8 vs. 3 µm	59
	C <sub>18</sub> Gravity, Sphinx RP, 5 µm	56
2-Acetylfuran	100-5 C <sub>18</sub> ec	59
4-Acetylpyridine	C <sub>18</sub> Isis, 5 µm	58
Acetylsalicylic acid	100-5 C <sub>8</sub> ec	33
	100-5 C <sub>18</sub> ec	32, 34
	C <sub>8</sub> Gravity, 5 µm	32
	C <sub>18</sub> Gravity, 5 µm	34
	C <sub>18</sub> HTec, 5 µm	23
	C <sub>18</sub> Pyramid, 5 µm	32
Acrylamide	HILIC, 5 µm	44
Adenine	100-5 NH <sub>2</sub> -RP	31
	C <sub>18</sub> Pyramid, 5 µm	47
	HILIC, 5 µm	27, 47
	PolarTec, 3 µm	17
	PolarTec, 5 µm	47
Adhumulone, adlupulone	100-5 C <sub>18</sub> ec	51
Adrenaline	C <sub>18</sub> Gravity, 5 µm	50
	HILIC, 3 µm	50
Aflatoxins	100-5 C <sub>18</sub> ec	50
Alanine	100-5 C <sub>18</sub> ec	46
4-Amino-2,6-dinitrotoluene	PolarTec, 5 µm	56
2-Amino-4,6-dinitrotoluene	PolarTec, 5 µm	56
4-Aminoanthraquinone	C <sub>18</sub> Isis, 5 µm	58
4-Aminobenzoic acid	C <sub>18</sub> Pyramid, 5 µm	52
p-Aminobenzoic acid	PolarTec, 5 µm	52
γ-Aminobutyric acid	100-5 C <sub>18</sub> ec	46
Aminomethylphosphonic acid	see AMPA	
Amitriptyline	C <sub>8</sub> Gravity, C <sub>18</sub> Isis, C <sub>18</sub> Pyramid	35
	C <sub>18</sub> Gravity, 5 µm	9, 35
	PolarTec, 5 µm	34
Amoxicillin	C <sub>18</sub> Gravity, 5 µm	42
	C <sub>18</sub> HTec, 5 µm	41
	C <sub>18</sub> Pyramid, 5 µm	40
	PFP, 5 µm	41
AMPA	100-5 NH <sub>2</sub> -RP	31, 60
	C <sub>18</sub> Gravity, 5 µm	60
Ampicillin	PFP, 5 µm	41
Aniline	C <sub>18</sub> HTec, 5 µm	22
p-Anisic acid	PolarTec, 3 µm	43
Anthracene	C <sub>18</sub> Gravity, 5 µm	59
	C <sub>18</sub> HTec, 5 µm; EC vs. VP	23
Arginine	100-5 C <sub>18</sub> ec	46
	C <sub>18</sub> Gravity, 3 µm	46
Ascorbic acid	100-5 C <sub>8</sub> ec	43
	C <sub>18</sub> Pyramid, 5 µm	36, 53
	HILIC, 3 µm	52
	HILIC, 5 µm	54
Asparagine	100-5 C <sub>18</sub> ec	46
Aspartame	100-5 C <sub>8</sub> ec	43
	C <sub>18</sub> HTec, 5 µm	43
Aspartic acid	100-5 C <sub>18</sub> ec	46
	100-5 CN-RP	54
Atrazine	100-3 C <sub>8</sub> ec	60, 61

Atropine	C <sub>8</sub> Gravity, 5 µm	48
Azorubine	C <sub>18</sub> Gravity, 5 µm	44

## B

Bambuterol	C <sub>18</sub> Pyramid, 5 µm	37
Beclometasone dipropionate	C <sub>18</sub> Pyramid, 1.8 µm	33
	100-3 C <sub>8</sub> ec	61
Bentazone	100-5 C <sub>18</sub> ec	58
Benzaldehyde	C <sub>18</sub> Gravity, Sphinx RP, 5 µm	56
	100-5 CN-RP	37
Benzalkonium chlorides	100-5 CN-RP	28, 29
Benzamide	C <sub>18</sub> Gravity, 5 µm	10, 15, 56
	C <sub>18</sub> Pyramid, C <sub>8</sub> Gravity, 5 µm	15
	PFP, 3 µm	57
	Sphinx RP, 5 µm	56
Benz[a]anthracene	C <sub>18</sub> Gravity, 5 µm	59
Benzocaine	C <sub>18</sub> Pyramid, 5 µm	32
Benzo[b]fluoranthene	C <sub>18</sub> Gravity, 5 µm	59
Benzo[k]fluoranthene	C <sub>18</sub> Gravity, 5 µm	59
Benzoic acid	100-5 C <sub>8</sub> ec	43
	C <sub>18</sub> Gravity, 5 µm	36
	C <sub>18</sub> HTec, 5 µm	43
	PolarTec, 3 µm	43
Benzo[ghi]perylene	C <sub>18</sub> Gravity, 5 µm	59
Benzophenone	100-5 C <sub>18</sub> ec	59
	C <sub>18</sub> Gravity, 1.8 vs. 3 µm	59
	C <sub>18</sub> Gravity, 5 µm	15, 56
	C <sub>18</sub> HTec, 5 µm	51
	C <sub>18</sub> Pyramid, C <sub>8</sub> Gravity, 5 µm	15
	Sphinx RP, 5 µm	56
Benzophenone-3	100-5 C <sub>18</sub> ec	51
Benzo[a]pyrene	C <sub>18</sub> Gravity, 5 µm	59
Benzyl alcohol	PolarTec, 3 µm	43
Betamethasone	C <sub>18</sub> Isis, 5 µm	51
	PFP, 5 µm	19
Biotin	C <sub>18</sub> Pyramid, 5 µm	53
	HILIC, 3 µm	52
Biphenyl	100-5 CN-RP	28, 29
	C <sub>18</sub> Pyramid, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	15
	Sphinx RP, 5 µm	62
	100-5 C <sub>8</sub> ec/C <sub>18</sub> ec	25
Biphenyl-2-ol	C <sub>18</sub> Pyramid, 5 µm	45
Biuret	100-3 C <sub>8</sub> ec	61
Bromacil	100-5 C <sub>18</sub> ec	38
Bromazepam	100-5 C <sub>18</sub> ec / 100-5 CN-RP	28
Brompheniramine	PFP, 5 µm	37
	PFP, C <sub>18</sub> Gravity, 5 µm	19
Brucine	C <sub>8</sub> Gravity, 5 µm	48
Butacaine	C <sub>18</sub> Pyramid, 5 µm	32
Butyl methoxydibenzoylmethane	100-5 C <sub>18</sub> ec	51
	PolarTec, 3 µm	43
Butyl paraben	100-5 C <sub>18</sub> ec	59
Butyrophenone	C <sub>18</sub> Gravity, 1.8 vs. 3 µm	59
<b>C</b>		
Caffeine	100-5 C <sub>8</sub> ec	43
	100-5 C <sub>18</sub> ec	24
	C <sub>8</sub> Gravity, 5 µm	32
	C <sub>18</sub> Gravity, 5 µm	11
	C <sub>18</sub> Isis, 5 µm	13
	C <sub>18</sub> Pyramid, 5 µm	36
	Sphinx RP, 5 µm	20, 42
4-Carboxybenzaldehyde	100-5 C <sub>18</sub> ec	58
Carprofen	100-5 C <sub>8</sub> ec/C <sub>18</sub> ec	33
Catechin	Sphinx RP, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	21
Catechol	PFP, 3 µm	57

NUCLEODUR®

# Alphabetical index of analytes

Cefalotin	PF	5 µm	41	Cytosine	100-5 NH <sub>2</sub> -RP	31
Cefamandole	C <sub>18</sub> Gravity, PFP,	5 µm	41		C <sub>18</sub> Pyramid, 5 µm	47
Cefotaxime	C <sub>18</sub> Gravity, PFP,	5 µm	41		HILIC, 5 µm	27, 47
Cefoxitin	C <sub>18</sub> Gravity, PFP,	5 µm	41		PolarTec, 3 µm	17
Cephalexin	PF	5 µm	41		PolarTec, 5 µm	47
Cephalothin	C <sub>18</sub> Gravity,	5 µm	41	<b>D</b>		
Cetirizine	PF	5 µm	37	2,4-D	100-3 C <sub>8</sub> ec	61
	PF, C <sub>18</sub> Gravity,	5 µm	19	Dehydroacetic acid	C <sub>18</sub> HTec, 5 µm	43
Chloramphenicol	C <sub>18</sub> Gravity,	5 µm	42		PolarTec, 3 µm	43
Chlormequat	HILIC,	3 µm	27	Dehydroascorbic acid	HILIC, 5 µm	54
4-Chloro-2-nitroaniline	PF	5 µm	58	Deoxycorticosterone	C <sub>18</sub> Gravity, 5 µm	49
4-Chloro-3-methylphenol	PF	5 µm	57	Desethylatrazine	100-3 C <sub>8</sub> ec	60, 61
Chloroaniline isomers	PF	5 µm	58		C <sub>18</sub> Isis, 5 µm	58
Chlorocresol	C <sub>18</sub> Pyramid,	1.8 µm	33	Desethylterbutylazine	100-3 C <sub>8</sub> ec	61
2-Chlorophenol	PF	5 µm	57	Desisopropylatrazine	100-3 C <sub>8</sub> ec	61
Chloroprothixene hydrochloride	C <sub>8</sub> Gravity,	5 µm	36	Dexamethasone	C <sub>18</sub> Gravity, 5 µm	49
	C <sub>18</sub> Gravity,	5 µm	48		C <sub>18</sub> Isis, 5 µm	51
Chloroxuron	100-3 C <sub>8</sub> ec		60		PF, 5 µm	19
Chlorpheniramine	100-5 C <sub>18</sub> ec / 100-5 CN-RP		28	Dextromethorphan	C <sub>18</sub> Gravity, 5 µm	36
	C <sub>8</sub> Gravity, 5 µm		15		C <sub>18</sub> Pyramid, 5 µm	36
	C <sub>18</sub> Gravity, 5 µm		15, 36	Dibenz[ah]anthracene	C <sub>18</sub> Gravity, 5 µm	59
	C <sub>18</sub> Pyramid, 5 µm		15, 36	Dibenzothiophene	100-5 NH <sub>2</sub>	30
	PF, 5 µm		37	Dibromophenol isomers	PF, 5 µm	57
	PF, C <sub>18</sub> Gravity, 5 µm		19		PF, C <sub>18</sub> HTec, 5 µm	19
Chlorpromazine	100-5 CN-RP		38	Dibutyl phthalate	C <sub>18</sub> Gravity, 5 µm	9
Chlortoluron	100-3 C <sub>8</sub> ec		60, 61	Dicamba	100-3 C <sub>8</sub> ec	61
Chrysene	C <sub>18</sub> Gravity, 5 µm		59	Dichlobenil	100-3 C <sub>8</sub> ec	60
Cimetidine	C <sub>18</sub> Gravity, 3 µm		36	3,4-Dichloroaniline	PF, 5 µm	58
Cinoxacin	100-5 C <sub>18</sub> ec		39	2,4-Dichlorobenzamide	100-3 C <sub>8</sub> ec	61
	C <sub>18</sub> Gravity, 5 µm		42	2,4-Dichlorophenol	PF, 5 µm	57
	C <sub>18</sub> Gravity, C <sub>8</sub> Gravity, C <sub>18</sub> Pyramid		40	Dichlorophenol isomers	PF, 5 µm	57
	C <sub>18</sub> HTec, 5 µm		41		PF, C <sub>18</sub> HTec, 5 µm	19
	Sphinx RP, 5 µm		40	Dichlorprop	100-3 C <sub>8</sub> ec	61
Ciprofloxacin	100-5 C <sub>18</sub> ec		39	Diclofenac	100-5 C <sub>8</sub> ec	33
	Sphinx RP, 5 µm		39		100-5 C <sub>18</sub> ec	32, 33, 34
Citric acid	C <sub>18</sub> Pyramid, 5 µm		55	Dicloxacillin	100-5 C <sub>18</sub> ec, C <sub>18</sub> Pyramid, 5 µm	40
	HILIC, 3 µm		55		PF, 5 µm	41
Citrulline	C <sub>18</sub> Gravity, 3 µm		46	Diethylenetrinitrilopentaacetic acid		
Clenbuterol	100-5 CN-RP		38		C <sub>18</sub> Pyramid, 5 µm	54
	C <sub>18</sub> Pyramid, 5 µm		37	Diflunisal	100-5 C <sub>8</sub> ec	33
Clobetasol 17-propionate	C <sub>18</sub> Pyramid, 1.8 µm		33		C <sub>18</sub> Gravity, 5 µm	34
Clomipramine	100-5 CN-RP; C <sub>18</sub> Isis, 5 µm		35	Dihydroxybenzoic acid isomers		
Clonidin	100-5 CN-RP		38		C <sub>18</sub> Isis, 5 µm	54
Cloxacillin	100-5 C <sub>18</sub> ec		39, 40		PolarTec, 5 µm	55
	C <sub>18</sub> Gravity, 5 µm		42	Dihydroxymandelic acid	PolarTec, 5 µm	55
	C <sub>18</sub> HTec, 5 µm		41	Dihydroxynaphthalene isomers		
	PF, 5 µm		41		C <sub>8</sub> Gravity, 5 µm	63
Cohumulone, colupulone	100-5 C <sub>18</sub> ec		51		C <sub>18</sub> Isis, 5 µm	63
Corticosterone	C <sub>18</sub> Gravity, 5 µm		49		Sphinx RP, 5 µm	62
Cortisone	100-5 C <sub>8</sub> ec		49	Dihydroxyphenylacetic acid		
	100-5 CN / CN-RP		29		PolarTec, 5 µm	55
	C <sub>8</sub> Gravity, 5 µm		48	Dihydroxyphenylalanine	C <sub>18</sub> Gravity, 5 µm	50
	C <sub>18</sub> Gravity, 5 µm		49	Dimefuron	100-3 C <sub>8</sub> ec	61
	C <sub>18</sub> HTec, 5 µm		49	<i>N,N</i> -Dimethylaniline	C <sub>18</sub> HTec, 5 µm	22
Creatine	HILIC, 3 µm		45		C <sub>18</sub> Isis, 5 µm	58
Creatinine	HILIC, 3 µm		45	1,2-Dimethylbenzene	100-5 NH <sub>2</sub>	30
<i>o</i> -, <i>m</i> -Cresol	PF, 5 µm		57	2,4-Dimethylphenol	PF, 5 µm	57
	PF, C <sub>18</sub> HTec, 5 µm		19	Dimethylphenol isomers	PF, 5 µm	57
Cyanazine	100-3 C <sub>8</sub> ec		60, 61		PF, C <sub>18</sub> HTec, 5 µm	19
Cyanocobalamine	C <sub>18</sub> Pyramid, 5 µm		53	Dimethyl phthalate	100-5 CN-RP	28, 29
	HILIC, 3 µm		52		C <sub>18</sub> Pyramid, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	15
Cyanuric acid	C <sub>18</sub> Pyramid, 5 µm		45	1,3-Dinitrobenzene	PolarTec, 5 µm	56
	HILIC, 5 µm		45	2,4-Dinitrophenol	PF, 5 µm	57
Cyclohexane	100-5 NH <sub>2</sub>		30	3,5-Dinitro-(1-phenylethylbenzamide)		
					C <sub>18</sub> Isis, 5 µm	58
				Dinitrotoluene isomers	PolarTec, 5 µm	56

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Diphenhydramine	C <sub>18</sub> Gravity, 5 µm	10	Fumaric acid	100-5 CN-RP	54
	C <sub>18</sub> Pyramid, 5 µm	36		C <sub>18</sub> Pyramid, 5 µm	55
	PFP, 5 µm	37		HILIC, 3 µm	55
	PFP, C <sub>18</sub> Gravity, 5 µm	19	2-Furfural	100-5 C <sub>18</sub> ec	59
Dipyridamole	C <sub>18</sub> Gravity, 3 µm	39	2-Furfuryl alcohol	100-5 C <sub>18</sub> ec	59
Diquat	HILIC, 3 µm	62			
Diuron	100-5 C <sub>8</sub> ec	60, 61	<b>G</b>		
L-DOPA	HILIC, 3 µm	50	Gallic acid	C <sub>18</sub> Isis, 5 µm	54
Dopamine	HILIC, 3 µm	50		C <sub>18</sub> Pyramid, 5 µm	43
Doxepin	100-5 CN-RP; C <sub>8</sub> Gravity, C <sub>18</sub> Gravity,			PolarTec, 5 µm	55
	C <sub>18</sub> Pyramid, 5 µm	35	Glucose	100-5 NH <sub>2</sub> -RP	30
	PolarTec, 5 µm	34	Glufosinate	100-5 NH <sub>2</sub> -RP	60
DTPA	C <sub>18</sub> Pyramid, 5 µm	54		C <sub>18</sub> Gravity, 5 µm	60
<b>E</b>			Glutamic acid	100-5 C <sub>18</sub> ec	46
E 102/104/110/122/124/126/127			Glutamine	100-5 C <sub>18</sub> ec	46
	C <sub>18</sub> Gravity, 5 µm	44		C <sub>18</sub> Gravity, 3 µm	46
EDTA	C <sub>18</sub> Pyramid, 5 µm	54	Glycine	100-5 C <sub>18</sub> ec	46
Enrofloxacin	C <sub>18</sub> Gravity, 5 µm	42	Glyphosate	100-5 NH <sub>2</sub> -RP	60
	C <sub>18</sub> HTec, 5 µm	41		C <sub>18</sub> Gravity, 5 µm	60
	Sphinx RP, 5 µm	39	Guanine	PolarTec, 3 µm	17
Ephedrine	100-5 C <sub>18</sub> ec/100-5 CN-RP	28		PolarTec, 5 µm	47
Erythrosine	C <sub>18</sub> Gravity, 5 µm	44	Guanosine	HILIC, 5 µm	27, 47
Estradiol	100-5 C <sub>8</sub> ec; C <sub>18</sub> HTec, 5 µm	49	<b>H</b>		
Estril	100-5 C <sub>8</sub> ec; C <sub>18</sub> HTec, 5 µm	49	Hexamethylbenzene	100-5 NH <sub>2</sub>	30
Estrone	100-5 C <sub>8</sub> ec; C <sub>18</sub> HTec, 5 µm	49	Hexazinone	100-3 C <sub>8</sub> ec	60, 61
2-Ethoxyphenol	100-5 C <sub>8</sub> ec	25	Hexobarbital	100-5 C <sub>18</sub> ec	34
	100-5 C <sub>18</sub> ec	25, 56	Hexogen (RDX)	PolarTec, 5 µm	56
4-Ethoxyvanillin	100-5 C <sub>18</sub> ec	58	Histamine	HILIC, 3 µm	47
4-Ethylaniline	C <sub>18</sub> HTec, 5 µm	22	Histidine	100-5 C <sub>18</sub> ec	46
	C <sub>18</sub> Isis, 5 µm	58		C <sub>18</sub> Gravity, 3 µm	46
Ethylbenzene	100-5 C <sub>18</sub> ec	24		HILIC, 3 µm	47
	C <sub>18</sub> Gravity, 1.8 vs. 3 µm	6	<i>n</i> -Humulone	PolarTec, 3 µm	17
	C <sub>18</sub> Gravity, 5 µm	11	Hydrocortisone	100-5 C <sub>18</sub> ec	51
	C <sub>18</sub> HTec, 5 µm	22		100-5 CN/CN-RP	29
	C <sub>18</sub> Isis, 5 µm	13		C <sub>8</sub> Gravity, 5 µm	48
Ethyl benzoate	C <sub>18</sub> Pyramid, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	15		C <sub>18</sub> Gravity, 5 µm	49
	Sphinx RP, 5 µm	62	Hydrocortisone acetate	C <sub>8</sub> Gravity, 5 µm	48
Ethylenediaminetetraacetic acid			4-Hydroxybenzaldehyde	100-5 C <sub>18</sub> ec	58
	C <sub>18</sub> Pyramid, 5 µm	54	4-Hydroxybenzoic acid	C <sub>18</sub> Isis, 5 µm	54
Ethylhexyldimethyl PABA	100-5 C <sub>18</sub> ec; C <sub>18</sub> HTec, 5 µm	51	3-Hydroxybenzoic acid	PolarTec, 5 µm	55
Ethylhexyl methoxycinnamate			17 $\alpha$ -Hydroxycortisone	C <sub>18</sub> Gravity, 5 µm	49
	100-5 C <sub>18</sub> ec; C <sub>18</sub> HTec, 5 µm	51	Hydroxymethylfurfural	C <sub>18</sub> Gravity, 5 µm	44
Ethylhexyl salicylate	100-5 C <sub>18</sub> ec; C <sub>18</sub> HTec, 5 µm	51	5-Hydroxymethyl-2-furfural	100-5 C <sub>18</sub> ec	59
Ethyl paraben	PolarTec, 3 µm	43	$\alpha$ -Hydroxymidazolam	C <sub>18</sub> Gravity, 3 µm	38
Eugenol	C <sub>18</sub> Gravity, 1.8 vs. 3 µm	59	4-Hydroxyproline	C <sub>18</sub> Gravity, 3 µm	46
<b>F</b>			Hydroxytestosterone isomers		
Famotidine	C <sub>18</sub> Gravity, 3 µm	36		C <sub>18</sub> Isis, 1.8 µm	49
Fast Red E	C <sub>18</sub> Gravity, 5 µm	44	Hydroxytyramine	C <sub>18</sub> Gravity, 5 µm	50
Fast Yellow	C <sub>18</sub> Gravity, 5 µm	44	Hydroxyzine	PFP, 5 µm	37
Fenoprofen	100-5 C <sub>8</sub> ec;	33		PFP, C <sub>18</sub> Gravity, 5 µm	19
	100-5 C <sub>18</sub> ec	33	<b>I</b>		
	C <sub>18</sub> Gravity, 5 µm	34	Ibuprofen	100-5 C <sub>8</sub> ec	33
Fenoterol	C <sub>18</sub> Pyramid, 5 µm	37		100-5 C <sub>18</sub> ec	32, 34
Fisetin	Sphinx RP, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	21		C <sub>18</sub> Gravity, 5 µm	34
Flumequine	Sphinx RP, 5 µm	39		C <sub>18</sub> Pyramid, 5 µm	32
Flunarizine hydrochloride	C <sub>18</sub> Gravity, 3 µm	39	Imipramine	C <sub>8</sub> Gravity, C <sub>18</sub> Gravity, C <sub>18</sub> Isis,	
Fluoranthene	C <sub>18</sub> Gravity, 5 µm	59		C <sub>18</sub> Pyramid, 5 µm	35
Fluorene	C <sub>18</sub> Gravity, 5 µm	59		PolarTec, 5 µm	34
Flurbiprofen	100-5 C <sub>8</sub> ec	33	Indeno[1,2,3-cd]pyrene	C <sub>18</sub> Gravity, 5 µm	59
	100-5 C <sub>18</sub> ec	34	Indomethacin	100-5 C <sub>8</sub> ec	33
	C <sub>18</sub> Gravity, 5 µm	34		C <sub>18</sub> Gravity, 5 µm	34
	C <sub>18</sub> Pyramid, 5 µm	32	Irgasan	PolarTec, 3 µm	43
Folic acid	C <sub>18</sub> Pyramid, 5 µm	52, 53	Isoadhumulone	100-5 C <sub>18</sub> ec	51
Formic acid	C <sub>18</sub> Pyramid, 5 µm	15	Isobutyl paraben	PolarTec, 3 µm	43
Fructose	100-5 NH <sub>2</sub> -RP	30	Isocohumulone	100-5 C <sub>18</sub> ec	51
			Isohumulone	100-5 C <sub>18</sub> ec	51

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Isoleucine	100-5 C <sub>18</sub> ec	46	6 $\alpha$ -Methyl-11 $\beta$ -hydroxyprogesterone	100-5 C <sub>8</sub> ec; C <sub>18</sub> HTec, 5 $\mu$ m	49
	C <sub>18</sub> Gravity, 3 $\mu$ m	46		C <sub>8</sub> Gravity, 5 $\mu$ m	48
Isoproturon	100-3 C <sub>8</sub> ec	60, 61	6 $\alpha$ -Methyl-17 $\alpha$ -hydroxyprogesterone / + acetate	100-5 C <sub>8</sub> ec; C <sub>18</sub> HTec, 5 $\mu$ m	49
Isorhamnetin	Sphinx RP, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	21		C <sub>8</sub> Gravity, 5 $\mu$ m	48
<b>K</b>			Methyl paraben	PolarTec, 3 $\mu$ m	43
Kaempferol	Sphinx RP, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	21	6 $\alpha$ -Methylprednisolone	C <sub>18</sub> Gravity, 5 $\mu$ m	49
Ketoprofen	100-5 C <sub>8</sub> ec	33	Methyltestosterone	100-5 CN / CN-RP	29
	100-5 C <sub>18</sub> ec	32, 33, 34	Metobromuron	100-3 C <sub>8</sub> ec	60, 61
	C <sub>18</sub> Gravity, 5 $\mu$ m	10	Metolachlor	100-3 C <sub>8</sub> ec	60, 61
	C <sub>18</sub> Pyramid, 5 $\mu$ m	32	Metoxuron	100-3 C <sub>8</sub> ec	60, 61
<b>L</b>			Midazolam	C <sub>18</sub> Gravity, 3 $\mu$ m	38
Lactic acid	C <sub>18</sub> Pyramid, 5 $\mu$ m	55	Monensin sodium	C <sub>18</sub> Gravity, 3 $\mu$ m	41
Lactose	100-5 NH <sub>2</sub> -RP	30	Monolinuron	100-3 C <sub>8</sub> ec	60, 61
Leucine	100-5 C <sub>18</sub> ec	46	<b>N</b>		
	C <sub>18</sub> Gravity, 3 $\mu$ m	46	Nafcillin	100-5 C <sub>18</sub> ec, C <sub>18</sub> Pyramid, 5 $\mu$ m	40
Lidocaine	C <sub>8</sub> Gravity, 5 $\mu$ m	15		PFM, 5 $\mu$ m	41
	C <sub>18</sub> Gravity, 5 $\mu$ m	10, 15	Nalidixic acid	C <sub>18</sub> Gravity, 5 $\mu$ m	42
	C <sub>18</sub> Pyramid, 5 $\mu$ m	15, 32		C <sub>18</sub> Gravity, C <sub>8</sub> Gravity, C <sub>18</sub> Pyramid	40
	Sphinx RP, 5 $\mu$ m	62		Sphinx RP, 5 $\mu$ m	40
Linuron	100-3 C <sub>8</sub> ec	60, 61		C <sub>18</sub> HTec, 5 $\mu$ m	41
Lorazepam	100-5 C <sub>18</sub> ec	38		Sphinx RP, 5 $\mu$ m	39
<i>n</i> -Lupulone	100-5 C <sub>18</sub> ec	51	Naphthalene	100-5 NH <sub>2</sub>	30
Lysine	100-5 C <sub>18</sub> ec	46		C <sub>18</sub> Gravity, 1.8 vs. 3 $\mu$ m	6
<b>M</b>				C <sub>18</sub> Gravity, 5 $\mu$ m	15, 59
Maleic acid	100-5 C <sub>18</sub> ec	28		C <sub>18</sub> HTec, 5 $\mu$ m; EC vs. VP	23
	100-5 CN-RP	28, 54	Naproxen	C <sub>18</sub> Pyramid, C <sub>8</sub> Gravity	15
	C <sub>18</sub> Gravity, 5 $\mu$ m	36		HILIC, 3 $\mu$ m	27
	C <sub>18</sub> Pyramid, 5 $\mu$ m	14		100-5 C <sub>8</sub> ec	33
	PFM, 5 $\mu$ m	37	Nicardipine	C <sub>18</sub> Gravity, 5 $\mu$ m	34
	PFM, C <sub>18</sub> Gravity, 5 $\mu$ m	19	Nicotinamide	100-5 CN-RP	39
Malic acid	C <sub>18</sub> Pyramid, 5 $\mu$ m	55		C <sub>18</sub> Pyramid, 5 $\mu$ m	52
Maltose	100-5 NH <sub>2</sub> -RP	30		HILIC, 3 $\mu$ m	52
Mapenterol	C <sub>18</sub> Pyramid, 5 $\mu$ m	37	Nicotinic acid	C <sub>18</sub> Pyramid, 5 $\mu$ m	52
Maprotiline	C <sub>18</sub> Isis, 5 $\mu$ m	35	Nifedipine	100-5 CN-RP	39
Marbofloxacin	Sphinx RP, 5 $\mu$ m	39		C <sub>18</sub> Gravity, 3 $\mu$ m	39
MCPA	100-3 C <sub>8</sub> ec	61	Niflumic acid	C <sub>18</sub> Gravity, 5 $\mu$ m	34
Meclofenamic acid	100-5 C <sub>18</sub> ec	34	Nimodipine	100-5 CN-RP	39
Mecoprop	100-3 C <sub>8</sub> ec	61	Nisoldipine	100-5 CN-RP	39
Medrysone	100-5 CN / CN-RP	29	Nitrendipine	100-5 CN-RP	39
Mefloquine	C <sub>18</sub> Gravity, 5 $\mu$ m	48	Nitrioltriacetic acid	C <sub>18</sub> Pyramid, 5 $\mu$ m	54
Melamine	HILIC, 5 $\mu$ m	45	Nitroaniline isomers	PFM, 5 $\mu$ m	58
Mephobarbital	100-5 C <sub>18</sub> ec	34	Nitrobenzene	C <sub>18</sub> Gravity, Sphinx RP, 5 $\mu$ m	56
Mepiquat	HILIC, 3 $\mu$ m	27		PFM, 3 $\mu$ m	57
Metalaxyl	100-3 C <sub>8</sub> ec	61		PolarTec, 5 $\mu$ m	56
Metamitron	100-3 C <sub>8</sub> ec	60	Nitrocresol isomers	C <sub>18</sub> Gravity, 5 $\mu$ m	56
Metazachlor	100-3 C <sub>8</sub> ec	60, 61	2-Nitrophenol	C <sub>18</sub> Gravity, Sphinx RP, 5 $\mu$ m	56
Methabenzthiazuron	100-3 C <sub>8</sub> ec	60, 61	Nitrophenol isomers	C <sub>18</sub> Gravity, 5 $\mu$ m	56
Methacrylamide	HILIC, 5 $\mu$ m	44		PFM, 5 $\mu$ m	57
Methacrylic acid	HILIC, 5 $\mu$ m	44	Nitrotoluene isomers	PolarTec, 5 $\mu$ m	56
Methionine	100-5 C <sub>18</sub> ec	46	Nizatidine	C <sub>18</sub> Gravity, 3 $\mu$ m	36
	C <sub>18</sub> Gravity, 3 $\mu$ m	46	<i>N</i> -Methyl- <i>N</i> -2,4,6-tetranitro-aniline (Tetryl)	PolarTec, 5 $\mu$ m	56
Methoxyphenol isomers	100-5 C <sub>8</sub> ec	25		100-5 C <sub>18</sub> ec / 100-5 CN-RP	28
2-Methyl-4,6-dinitrophenol	PFM, 5 $\mu$ m	57	Norephedrine	C <sub>18</sub> Gravity, 5 $\mu$ m	50
<i>o</i> -, <i>m</i> -, <i>p</i> -Methylacetophenone	PFM, C <sub>18</sub> Gravity, 5 $\mu$ m	58		HILIC, 3 $\mu$ m	50
9-Methylantracene	100-5 NH <sub>2</sub>	30	Norgestrel	100-5 CN / CN-RP	29
4-Methylbenzylidene camphor	100-5 C <sub>18</sub> ec	51	Nortriptyline	100-5 CN-RP; C <sub>8</sub> Gravity, C <sub>18</sub> Gravity,	
	C <sub>18</sub> HTec, 5 $\mu$ m	51		C <sub>18</sub> Isis, C <sub>18</sub> Pyramid, 5 $\mu$ m	35
5-Methyl-2-furfurol	100-5 C <sub>18</sub> ec	59	Norvaline	100-5 C <sub>18</sub> ec	46
Methylhistidine isomers	PolarTec, 3 $\mu$ m	17	Noscapine	C <sub>8</sub> Gravity, 5 $\mu$ m	48
Methyl-4-hydroxybenzoate	C <sub>18</sub> Pyramid, 5 $\mu$ m	32		C <sub>18</sub> Gravity, 5 $\mu$ m	10
			NTA	C <sub>18</sub> Pyramid, 5 $\mu$ m	54

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## O

Octocrylene	100-5 C <sub>18</sub> ec; C <sub>18</sub> HTec, 5 µm	51
Octogen (HMX)	PolarTec, 5 µm	56
Ofloxacin	100-5 C <sub>18</sub> ec	39
Ornithine	C <sub>18</sub> Gravity, 3 µm	46
Oxacillin	PFP, 5 µm	41
Oxalic acid	HILIC, 3 µm	55
Oxazepam	100-5 C <sub>18</sub> ec	38
Oxolinic acid	C <sub>8</sub> Gravity, C <sub>18</sub> Pyramid, 5 µm	40
	C <sub>18</sub> Gravity, 5 µm	40, 42
	C <sub>18</sub> HTec, 5 µm	41
	Sphinx RP, 5 µm	39, 40

## P

Panhotenic acid	PolarTec, 5 µm	52
Papaverine	C <sub>8</sub> Gravity, 5 µm	48
	C <sub>18</sub> Gravity, 5 µm	10
Paracetamol	100-5 C <sub>18</sub> ec	28, 32
	100-5 CN-RP	28
	C <sub>8</sub> Gravity, 5 µm	32
	C <sub>18</sub> Gravity, 5 µm	36
	C <sub>18</sub> Pyramid, 5 µm	32, 36
Paraquat	HILIC, 3 µm	62
Patulin	C <sub>18</sub> Gravity, 5 µm	44
Penicillins G + V	100-5 C <sub>18</sub> ec	39, 40
	C <sub>18</sub> Pyramid, 5 µm	40
Penicillin V	C <sub>18</sub> Gravity, 5 µm	42
	C <sub>18</sub> HTec, PFP, 5 µm	41
Pentachlorophenol	PFP, 5 µm	57
Pentobarbital	100-5 C <sub>18</sub> ec	34
Perfluorinated surfactants	Sphinx RP, 3 µm	62
Phenactin	C <sub>8</sub> Gravity, 5 µm	32
Phenanthrene	C <sub>18</sub> Gravity, 5 µm	59
Phenetole	100-5 C <sub>8</sub> ec / C <sub>18</sub> ec	25
	100-5 CN-RP	28, 29
Phenobarbital	100-5 C <sub>18</sub> ec	34
Phenol	100-5 C <sub>8</sub> ec	25
	100-5 C <sub>18</sub> ec	25, 56
	C <sub>18</sub> Gravity, 5 µm	56
	C <sub>18</sub> HTec, 5 µm	22
	C <sub>18</sub> HTec, 5 µm; EC vs. VP	23
	PFP, 3 µm	57
	PFP, 5 µm	57
	PolarTec, 5 µm	17
	Sphinx RP, 5 µm	21, 56
Phenoxyethanol	PolarTec, 3 µm	43
Phenylalanine	100-5 C <sub>18</sub> ec; C <sub>18</sub> Gravity, 3 µm	46
	HILIC, 3 µm	47
1-Phenyldodecane	100-5 NH <sub>2</sub>	30
Phenylglycine	HILIC, 3 µm	47
Phloroglucinol	PFP, 3 µm	57
Piperacillin	PFP, 5 µm	41
Pirenzepine hydrochloride	C <sub>18</sub> Gravity, 3 µm	36
Piroxicam	100-5 C <sub>8</sub> ec, 100-5 C <sub>18</sub> ec	33
	C <sub>18</sub> Gravity, 5 µm	34
Ponceau 4R and 6R	C <sub>18</sub> Gravity, 5 µm	44
Prednisolone	100-5 C <sub>8</sub> ec	49
	100-5 CN / CN-RP	29
	C <sub>18</sub> Gravity, 5 µm	49
	C <sub>18</sub> HTec, 5 µm	49
Procainamide	100-5 CN-RP	38
Progesterone	100-5 C <sub>8</sub> ec; C <sub>18</sub> Gravity,	
	C <sub>18</sub> HTec, 5 µm	49
Promazine	100-5 CN-RP	38
Promethazine	100-5 CN-RP	38
	PFP, 5 µm	37
	PFP, C <sub>18</sub> Gravity, 5 µm	19

Propazine	100-3 C <sub>8</sub> ec	61
Propiophenone	100-5 C <sub>18</sub> ec	59
	C <sub>18</sub> Gravity, 1.8 vs. 3 µm	59
Propyl 4-hydroxybenzoate	C <sub>18</sub> Gravity, Sphinx RP, 5 µm	56
Propyl paraben	PolarTec, 3 µm	43
Propyzamid	100-3 C <sub>8</sub> ec	60
Protriptyline	C <sub>8</sub> Gravity, C <sub>18</sub> Gravity, C <sub>18</sub> Isis,	
	C <sub>18</sub> Pyramid, 5 µm	35
Protryptiline	PolarTec, 5 µm	34
Pseudoephedrine	C <sub>18</sub> Gravity, 5 µm	36
Pyrene	C <sub>18</sub> Gravity, 5 µm	59
Pyridine	C <sub>18</sub> Gravity, 5 µm	11
	C <sub>18</sub> Isis, 5 µm	13, 58
	PolarTec, 5 µm	17
	Sphinx RP, 5 µm	21
Pyridoxal	C <sub>18</sub> Pyramid, 5 µm	53
Pyridoxamine	C <sub>18</sub> Pyramid, 5 µm	53
Pyridoxine	C <sub>18</sub> Pyramid, 5 µm	53
	HILIC, 3 µm	52
Pyridoxine hydrochloride	C <sub>18</sub> Pyramid, 5 µm	52
Pyrocatechol	100-5 C <sub>8</sub> ec	25
	100-5 C <sub>18</sub> ec	25, 56
Pyrogallol	PFP, 3 µm	57
<b>Q</b>		
Quercetin	Sphinx RP, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	21
Quinine	100-5 C <sub>8</sub> ec	43
	C <sub>8</sub> Gravity, C <sub>18</sub> Gravity, 5 µm	48
	C <sub>18</sub> Gravity, 5 µm	44
Quinoline yellow		
<b>R</b>		
Ractopamine	C <sub>18</sub> Pyramid, 5 µm	37
Reproterol	C <sub>18</sub> Pyramid, 5 µm	37
Resorcinol	100-5 C <sub>8</sub> ec	25
	100-5 C <sub>18</sub> ec	25, 56
	PFP, 3 µm	57
Riboflavin	C <sub>18</sub> Pyramid, 5 µm	52, 53
Ritodrine	C <sub>18</sub> Pyramid, 5 µm	37
Rutin	C <sub>18</sub> Pyramid, 5 µm	52
	Sphinx RP, C <sub>18</sub> Gravity, C <sub>8</sub> Gravity	21
<b>S</b>		
Saccharin	100-5 C <sub>8</sub> ec; C <sub>18</sub> HTec, 5 µm	43
Saccharose	100-5 NH <sub>2</sub> -RP	30
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## Trademarks

### MACHEREY-NAGEL trademarks

<b>ALUGRAM</b>	coated aluminium sheets for TLC
<b>CHROMABOND</b>	columns for solid phase extraction (SPE)
<b>ChromCart</b>	cartridge system for HPLC
<b>NUCLEODUR</b>	spherical high purity silica for HPLC

### Registered trademarks of other companies

Acclaim	Dionex Corp. (USA)
ACQUITY	Waters Corp. (USA)
Allure	Restek Corp. (USA)
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HyPURITY	Thermo Fisher Scientific Inc. (USA)
Obelisc	Sielc Technologies (USA)
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### NUCLEOSIL OPTIMA

spherical standard silica for HPLC  
high performance fused silica capillary  
columns with immobilized phases

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Xterra	Waters Corp. (USA)
YMC	YMC Co. Ltd. (Japan)
ZIC	Merck SeQuant AB (Sweden)
Zorbax	Agilent Technologies Inc. (USA)

SymmetryShield	Waters Corp. (USA)
SunFire	Waters Corp. (USA)
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## NUCLEODUR® C<sub>18</sub> Gravity

Pore size 110 Å; high density octadecyl phase, endcapped, 18% C;  
eluent in column acetonitrile - water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® C<sub>18</sub> Gravity, 1.8 µm</b>							particle size 1.8 µm
EC analytical columns							
2 mm ID	760078.20	760079.20	760071.20	760076.20		760075.20	
3 mm ID	760078.30	760079.30		760076.30			
4 mm ID	760078.40	760079.40		760076.40			
4.6 mm ID	760078.46	760079.46		760076.46			
EC guard columns*		4 x 2 mm: 761901.20		4 x 3 mm: 761901.30			
<b>NUCLEODUR® C<sub>18</sub> Gravity, 3 µm</b>							particle size 3 µm
Microbore analytical columns							
1 mm ID				717714.10	717715.10	717716.10	717717.10
EC analytical columns							
2 mm ID		760080.20		760084.20	760081.20	760083.20	760082.20
3 mm ID		760080.30		760084.30	760081.30	760083.30	760082.30
4 mm ID		760080.40		760084.40	760081.40	760083.40	760082.40
4.6 mm ID		760080.46	760086.46	760084.46	760081.46	760083.46	760082.46
EC guard columns*		4 x 2 mm: 761902.20		4 x 3 mm: 761902.30			
CC guard columns**		8 x 3 mm: 761124.30		8 x 4 mm: 761124.40			
<b>NUCLEODUR® C<sub>18</sub> Gravity, 5 µm</b>							particle size 5 µm
Microbore analytical columns							
1 mm ID				717706.10	717707.10	717708.10	717705.10
EC analytical columns							
2 mm ID		760102.20		760104.20	760100.20	760103.20	760101.20
3 mm ID		760102.30		760104.30	760100.30	760103.30	760101.30
4 mm ID		760102.40		760104.40	760100.40	760103.40	760101.40
4.6 mm ID		760102.46	760106.46	760104.46	760100.46	760103.46	760101.46
EC guard columns*		4 x 2 mm: 761903.20		4 x 3 mm: 761903.30			
CC guard columns**		8 x 3 mm: 761125.30		8 x 4 mm: 761125.40			
VarioPrep preparative columns							
10 mm ID		762103.100			762109.100		762113.100
21 mm ID		762103.210			762109.210		762113.210
32 mm ID							762113.320
40 mm ID						762100.400	762113.400
VP guard columns***		10 x 8 mm: 762160.80		10 x 16 mm: 762160.160		15 x 32 mm: 762163.320	
<b>NUCLEODUR® C<sub>18</sub> Gravity, 10 µm</b>							particle size 10 µm
VarioPrep preparative columns							
21 mm ID							762250.210
40 mm ID							762250.400
VP guard columns***		10 x 8 mm: 762160.80		10 x 16 mm: 762160.160		15 x 32 mm: 762163.320	
Microbore, EC, and VarioPrep columns in packs of 1, guard columns see page 73							

# Packed columns

## NUCLEODUR® C<sub>8</sub> Gravity

Pore size 110 Å; high density octyl phase, endcapped, 11% C;  
eluent in column acetonitrile – water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® C<sub>8</sub> Gravity, 1.8 µm</b>							particle size 1.8 µm
<b>EC analytical columns</b>							
2 mm ID	760756.20	760755.20	760760.20	760757.20		760759.20	
3 mm ID	760756.30	760755.30		760757.30			
4 mm ID	760756.40	760755.40		760757.40			
4.6 mm ID	760756.46	760755.46		760757.46			
EC guard columns*		4 x 2 mm: 761905.20		4 x 3 mm: 761905.30			
<b>NUCLEODUR® C<sub>8</sub> Gravity, 5 µm</b>							particle size 5 µm
<b>EC analytical columns</b>							
2 mm ID		760750.20		760754.20	760751.20	760752.20	760753.20
3 mm ID		760750.30		760754.30	760751.30	760752.30	760753.30
4 mm ID		760750.40		760754.40	760751.40	760752.40	760753.40
4.6 mm ID		760750.46	760749.46	760754.46	760751.46	760752.46	760753.46
EC guard columns*		4 x 2 mm: 761907.20		4 x 3 mm: 761907.30			
CC guard columns**		8 x 3 mm: 761754.30		8 x 4 mm: 761754.40			
<b>VarioPrep preparative columns</b>							
10 mm ID		762081.100			762071.100		762070.100
21 mm ID		762081.210			762071.210	762082.210	762070.210
VP guard columns***		10 x 8 mm: 762097.80		10 x 16 mm: 762097.160			
EC and VarioPrep columns in packs of 1, guard columns see right, Microbore columns with NUCLEODUR® C <sub>8</sub> Gravity on request!							

## HPLC column systems from MACHERY-NAGEL



microbore column



EC column



VarioPrep column

**Microbore columns:** On request available in lengths of 40, 60, 100, 125, 150, 200, 250 and 300 mm and with 0.05, 0.075, 0.1, 0.15, 0.3, 0.4, 0.5, 0.75, 1.0 and 1.5 mm ID.

**EC columns:** Analytical ready-to-use columns; available dimensions see page 85.

**VarioPrep columns:** Preparative columns with axially adjustable endfitting; available dimensions see page 88.

## NUCLEODUR® C<sub>18</sub> Isis

Pore size 110 Å; octadecyl phase with high steric selectivity, polymer modification, 20% C; eluent in column acetonitrile – water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® C<sub>18</sub> Isis, 1.8 µm</b>							particle size 1.8 µm
<b>EC analytical columns</b>							
2 mm ID	760406.20	760405.20	760396.20	760407.20		760409.20	
3 mm ID	760406.30	760405.30		760407.30			
4 mm ID	760406.40	760405.40		760407.40			
4.6 mm ID	760406.46	760405.46		760407.46			
EC guard columns*	4 x 2 mm: 761910.20		4 x 3 mm: 761910.30				
<b>NUCLEODUR® C<sub>18</sub> Isis, 3 µm</b>							particle size 3 µm
<b>Microbore analytical columns</b>							
1 mm ID		717760.10		717761.10	717762.10		
<b>EC analytical columns</b>							
2 mm ID		760400.20		760401.20	760402.20	760403.20	760404.20
3 mm ID		760400.30		760401.30	760402.30	760403.30	760404.30
4 mm ID		760400.40		760401.40	760402.40	760403.40	760404.40
4.6 mm ID		760400.46	760397.46	760401.46	760402.46	760403.46	760404.46
EC guard columns*		4 x 2 mm: 761911.20		4 x 3 mm: 761911.30			
CC guard columns**		8 x 3 mm: 761300.30		8 x 4 mm: 761300.40			
<b>NUCLEODUR® C<sub>18</sub> Isis, 5 µm</b>							particle size 5 µm
<b>Microbore analytical columns</b>							
1 mm ID		717770.10		717771.10	717772.10		
<b>EC analytical columns</b>							
2 mm ID		760410.20		760415.20	760412.20	760413.20	760414.20
3 mm ID		760410.30		760415.30	760412.30	760413.30	760414.30
4 mm ID		760410.40		760415.40	760412.40	760413.40	760414.40
4.6 mm ID		760410.46	760416.46	760415.46	760412.46	760413.46	760414.46
EC guard columns*		4 x 2 mm: 761912.20		4 x 3 mm: 761912.30			
CC guard columns**		8 x 3 mm: 761310.30		8 x 4 mm: 761310.40			
<b>VarioPrep preparative columns</b>							
10 mm ID		762404.100			762405.100		762403.100
21 mm ID		762404.210			762405.210		762403.210
32 mm ID							762403.320
40 mm ID						762406.400	762403.400
VP guard columns***		10 x 8 mm: 762420.80		10 x 16 mm: 762420.160		15 x 32 mm: 762422.320	

Microbore, EC, and VarioPrep columns in packs of 1, guard columns see below

### Guard column systems

Guard columns for EC columns with ID		2 mm	3 mm	4 mm	4.6 mm	Guard column holder	
*	Column Protection System (pack of)	EC	4/2 (3)	4/3 (3)	4/3 (3)	4/3 (3)	718966
**	ChromCart® guard columns (pack of)	CC	8/3 (3)	8/3 (3)	8/4 (3)	8/4 (3)	721359
Guard columns for VarioPrep columns with ID		8, 10 mm	16, 21 mm	32, 40 mm	≥ 50 mm		
***	VarioPrep guard columns (pack of)	VP	10/8 (2)	10/16 (2)	15/32 (1)	15/50 (1)	
	VP guard column holder		718251	718256	718253	718255	

# Packed columns

## NUCLEODUR® C<sub>18</sub> Pyramid

Pore size 110 Å; octadecyl phase with hydrophilic endcapping, 14% C; eluent in column acetonitrile - water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® C<sub>18</sub> Pyramid, 1.8 µm</b>							particle size 1.8 µm
EC analytical columns							
2 mm ID	760271.20	760272.20	760275.20	760273.20		760274.20	
3 mm ID	760271.30	760272.30		760273.30			
4 mm ID	760271.40	760272.40		760273.40			
4.6 mm ID	760271.46	760272.46		760273.46			
EC guard columns*	4 x 2 mm: 761915.20		4 x 3 mm: 761915.30				
<b>NUCLEODUR® C<sub>18</sub> Pyramid, 3 µm</b>							particle size 3 µm
Microbore analytical columns							
1 mm ID		717740.10		717741.10	717742.10	717743.10	717744.10
EC analytical columns							
2 mm ID		760263.20		760264.20	760260.20	760261.20	760262.20
3 mm ID		760263.30		760264.30	760260.30	760261.30	760262.30
4 mm ID		760263.40		760264.40	760260.40	760261.40	760262.40
4.6 mm ID		760263.46	760259.46	760264.46	760260.46	760261.46	760262.46
EC guard columns*	4 x 2 mm: 761916.20		4 x 3 mm: 761916.30				
CC guard columns**	8 x 3 mm: 761854.30		8 x 4 mm: 761854.40				
<b>NUCLEODUR® C<sub>18</sub> Pyramid, 5 µm</b>							particle size 5 µm
Microbore analytical columns							
1 mm ID				717722.10	717723.10	717724.10	717725.10
EC analytical columns							
2 mm ID		760200.20		760204.20	760201.20	760203.20	760202.20
3 mm ID		760200.30		760204.30	760201.30	760203.30	760202.30
4 mm ID		760200.40		760204.40	760201.40	760203.40	760202.40
4.6 mm ID		760200.46	760205.46	760204.46	760201.46	760203.46	760202.46
EC guard columns*	4 x 2 mm: 761917.20		4 x 3 mm: 761917.30				
CC guard columns**	8 x 3 mm: 761800.30		8 x 4 mm: 761800.40				
VarioPrep preparative columns							
10 mm ID		762271.100			762273.100		762272.100
21 mm ID		762271.210			762273.210		762272.210
32 mm ID							762272.320
40 mm ID						762269.400	762272.400
VP guard columns***	10 x 8 mm: 762291.80		10 x 16 mm: 762291.160		15 x 32 mm: 762293.320		
Microbore, EC, and VarioPrep columns in packs of 1, guard columns see right							

## NUCLEODUR® PolarTec

Pore size 110 Å; octadecyl phase with embedded polar group, endcapped, 17% C;  
eluent in column acetonitrile - water

Length →	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® PolarTec, 3 µm</b>						particle size 3 µm
<b>EC analytical columns</b>						
2 mm ID	760473.20		760476.20	760477.20	760478.20	760479.20
3 mm ID	760473.30		760476.30	760477.30	760478.30	760479.30
4 mm ID	760473.40		760476.40	760477.40	760478.40	760479.40
4.6 mm ID	760473.46	760475.46	760476.46	760477.46	760478.46	760479.46
EC guard columns*		4 x 2 mm: 761981.20		4 x 3 mm: 761981.30		
CC guard columns**		8 x 3 mm: 761160.30		8 x 4 mm: 761160.40		
<b>NUCLEODUR® PolarTec, 5 µm</b>						particle size 5 µm
<b>EC analytical columns</b>						
2 mm ID	760483.20		760486.20	760487.20	760488.20	760489.20
3 mm ID	760483.30		760486.30	760487.30	760488.30	760489.30
4 mm ID	760483.40		760486.40	760487.40	760488.40	760489.40
4.6 mm ID	760483.46	760485.46	760486.46	760487.46	760488.46	760489.46
EC guard columns*		4 x 2 mm: 761982.20		4 x 3 mm: 761982.30		
CC guard columns**		8 x 3 mm: 761161.30		8 x 4 mm: 761161.40		
<b>VarioPrep preparative columns</b>						
10 mm ID	762220.100			762221.100		762223.100
21 mm ID	762220.210			762221.210		762223.210
32 mm ID						762223.320
40 mm ID					762222.400	762223.400
VP guard columns***	10 x 8 mm: 762224.80		10 x 16 mm: 762224.160		15 x 32 mm: 762226.320	
EC and VarioPrep columns in packs of 1, guard columns see below						

Guard column systems							Guard column holder
Guard columns for EC columns with ID			2 mm	3 mm	4 mm	4.6 mm	
*	Column Protection System (pack of)	EC	4/2 (3)	4/3 (3)	4/3 (3)	4/3 (3)	718966
**	ChromCart® guard columns (pack of)	CC	8/3 (3)	8/3 (3)	8/4 (3)	8/4 (3)	721359
Guard columns for VarioPrep columns with ID			8, 10 mm	16, 21 mm	32, 40 mm	≥ 50 mm	
***	VarioPrep guard columns (pack of)	VP	10/8 (2)	10/16 (2)	15/32 (1)	15/50 (1)	
	VP guard column holder		718251	718256	718253	718255	

# Packed columns

## NUCLEODUR® PFP

Pore size 110 Å; pentafluorophenyl-propyl modification, multi-encapped, 8% C; eluent in column acetonitrile – water

Length →	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® PFP, 3 µm</b>						particle size 3 µm
<b>EC analytical columns</b>						
2 mm ID	760443.20		760446.20	760447.20	760448.20	760449.20
3 mm ID	760443.30		760446.30	760447.30	760448.30	760449.30
4 mm ID	760443.40		760446.40	760447.40	760448.40	760449.40
4.6 mm ID	760443.46	760445.46	760446.46	760447.46	760448.46	760449.46
EC guard columns*		4 x 2 mm: 761976.20		4 x 3 mm: 761976.30		
CC guard columns**		8 x 3 mm: 761145.30		8 x 4 mm: 761145.40		
<b>NUCLEODUR® PFP, 5 µm</b>						particle size 5 µm
<b>EC analytical columns</b>						
2 mm ID	760453.20		760456.20	760457.20	760458.20	760459.20
3 mm ID	760453.30		760456.30	760457.30	760458.30	760459.30
4 mm ID	760453.40		760456.40	760457.40	760458.40	760459.40
4.6 mm ID	760453.46	760455.46	760456.46	760457.46	760458.46	760459.46
EC guard columns*		4 x 2 mm: 761977.20		4 x 3 mm: 761977.30		
CC guard columns**		8 x 3 mm: 761146.30		8 x 4 mm: 761146.40		
<b>VarioPrep preparative columns</b>						
10 mm ID	762210.100			762211.100		762213.100
21 mm ID	762210.210			762211.210		762213.210
32 mm ID						762213.320
40 mm ID					762212.400	762213.400
VP guard columns***	10 x 8 mm: 762214.80		10 x 16 mm: 762214.160		15 x 32 mm: 762216.320	
EC and VarioPrep columns in packs of 1, guard columns see right						

## Online Application Database • [www.mn-net.com/apps](http://www.mn-net.com/apps)

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## NUCLEODUR® Sphinx RP

Pore size 110 Å; special bifunctional RP phase, 15% C;  
eluent in column acetonitrile – water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® Sphinx RP, 1.8 µm</b>							particle size 1.8 µm
EC analytical columns							
2 mm ID	760821.20	760822.20	760825.20	760823.20		760824.20	
3 mm ID	760821.30	760822.30		760823.30			
4 mm ID	760821.40	760822.40		760823.40			
4.6 mm ID	760821.46	760822.46		760823.46			
EC guard columns*	4 x 2 mm: 761920.20		4 x 3 mm: 761920.30				
<b>NUCLEODUR® Sphinx RP, 3 µm</b>							particle size 3 µm
EC analytical columns							
2 mm ID		760806.20		760812.20	760807.20	760805.20	760808.20
3 mm ID		760806.30		760812.30	760807.30	760805.30	760808.30
4 mm ID		760806.40		760812.40	760807.40	760805.40	760808.40
4.6 mm ID		760806.46	760813.46	760812.46	760807.46	760805.46	760808.46
EC guard columns*	4 x 2 mm: 761921.20		4 x 3 mm: 761921.30				
CC guard columns**	8 x 3 mm: 761557.30		8 x 4 mm: 761557.40				
<b>NUCLEODUR® Sphinx RP, 5 µm</b>							particle size 5 µm
Microbore analytical columns							
1 mm ID		717680.10		717681.10	717682.10	717683.10	717684.10
EC analytical columns							
2 mm ID		760800.20		760809.20	760801.20	760802.20	760803.20
3 mm ID		760800.30		760809.30	760801.30	760802.30	760803.30
4 mm ID		760800.40		760809.40	760801.40	760802.40	760803.40
4.6 mm ID		760800.46	760815.46	760809.46	760801.46	760802.46	760803.46
EC guard columns*	4 x 2 mm: 761922.20		4 x 3 mm: 761922.30				
CC guard columns**	8 x 3 mm: 761550.30		8 x 4 mm: 761550.40				
VarioPrep preparative columns							
10 mm ID		762372.100			762375.100		762373.100
21 mm ID		762372.210			762375.210		762373.210
32 mm ID							762373.320
40 mm ID						762371.400	762373.400
VP guard columns***	10 x 8 mm: 762390.80		10 x 16 mm: 762390.160		15 x 32 mm: 762392.320		

Microbore, EC, and VarioPrep columns in packs of 1, guard columns see below

Guard column systems							Guard column holder
Guard columns for EC columns with ID			2 mm	3 mm	4 mm	4.6 mm	
*	Column Protection System (pack of)	EC	4/2 (3)	4/3 (3)	4/3 (3)	4/3 (3)	718966
**	ChromCart® guard columns (pack of)	CC	8/3 (3)	8/3 (3)	8/4 (3)	8/4 (3)	721359
Guard columns for VarioPrep columns with ID			8, 10 mm	16, 21 mm	32, 40 mm	≥ 50 mm	
***	VarioPrep guard columns (pack of)	VP	10/8 (2)	10/16 (2)	15/32 (1)	15/50 (1)	
	VP guard column holder		718251	718256	718253	718255	

# Packed columns

## NUCLEODUR® C<sub>18</sub> HTec

Pore size 110 Å; high density octadecyl phase, endcapped, 18% C;  
eluent in column acetonitrile - water

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® C<sub>18</sub> HTec, 1.8 µm</b>							particle size 1.8 µm
EC analytical columns							
2 mm ID	760301.20	760305.20	760304.20	760306.20		760308.20	
3 mm ID	760301.30	760305.30		760306.30			
4 mm ID	760301.40	760305.40		760306.40			
4.6 mm ID	760301.46	760305.46		760306.46			
EC guard columns*	4 x 2 mm: 761925.20		4 x 3 mm: 761925.30				
<b>NUCLEODUR® C<sub>18</sub> HTec, 3 µm</b>							particle size 3 µm
EC analytical columns							
2 mm ID		760321.20		760323.20	760324.20	760325.20	760326.20
3 mm ID		760321.30		760323.30	760324.30	760325.30	760326.30
4 mm ID		760321.40		760323.40	760324.40	760325.40	760326.40
4.6 mm ID		760321.46	760322.46	760323.46	760324.46	760325.46	760326.46
EC guard columns*	4 x 2 mm: 761926.20		4 x 3 mm: 761926.30				
CC guard columns**	8 x 3 mm: 761120.30		8 x 4 mm: 761120.40				
<b>NUCLEODUR® C<sub>18</sub> HTec, 5 µm</b>							particle size 5 µm
EC analytical columns							
2 mm ID		760311.20		760313.20	760314.20	760315.20	760316.20
3 mm ID		760311.30		760313.30	760314.30	760315.30	760316.30
4 mm ID		760311.40		760313.40	760314.40	760315.40	760316.40
4.6 mm ID		760311.46	760312.46	760313.46	760314.46	760315.46	760316.46
EC guard columns*	4 x 2 mm: 761927.20		4 x 3 mm: 761927.30				
CC guard columns**	8 x 3 mm: 761110.30		8 x 4 mm: 761110.40				
VarioPrep preparative columns							
10 mm ID		762551.100			762554.100		762556.100
21 mm ID		762551.210		762553.210	762554.210		762556.210
32 mm ID				762553.320		762555.320	762556.320
40 mm ID						762555.400	762556.400
50 mm ID				762553.500		762555.500	762556.500
VP guard columns***	10 x 8 mm: 762591.80		10 x 16 mm: 762591.160				
	15 x 32 mm: 762592.320		15 x 50 mm: 762592.500				
<b>NUCLEODUR® C<sub>18</sub> HTec, 7 µm</b>							particle size 7 µm
VarioPrep preparative columns							
10 mm ID		762561.100			762564.100		762566.100
21 mm ID		762561.210		762563.210	762564.210		762566.210
32 mm ID				762563.320		762565.320	762566.320
40 mm ID						762565.400	762566.400
50 mm ID				762563.500		762565.500	762566.100
VP guard columns***	10 x 8 mm: 762591.80		10 x 16 mm: 762591.160				
	15 x 32 mm: 762592.320		15 x 50 mm: 762592.500				












## Ordering information

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® C<sub>18</sub> HTec, 10 µm</b>							particle size 10 µm
<b>VarioPrep preparative columns</b>							
10 mm ID		762571.100			762574.100		762576.100
21 mm ID		762571.210		762573.210	762574.210		762576.210
32 mm ID				762573.320		762575.320	762576.320
40 mm ID						762575.400	762576.400
50 mm ID				762573.500		762575.500	762576.100
VP guard columns***		10 x 8 mm: 762591.80		10 x 16 mm: 762591.160			
		15 x 32 mm: 762592.320		15 x 50 mm: 762592.500			

EC and VarioPrep columns in packs of 1, guard columns see below

Guard column systems							Guard column holder
Guard columns for EC columns with ID			2 mm	3 mm	4 mm	4.6 mm	
* Column Protection System (pack of)	EC		4/2 (3)	4/3 (3)	4/3 (3)	4/3 (3)	718966
** ChromCart® guard columns (pack of)	CC		8/3 (3)	8/3 (3)	8/4 (3)	8/4 (3)	721359
Guard columns for VarioPrep columns with ID			8, 10 mm	16, 21 mm	32, 40 mm	≥ 50 mm	
*** VarioPrep guard columns (pack of)	VP		10/8 (2)	10/16 (2)	15/32 (1)	15/50 (1)	
VP guard column holder			718251	718256	718253	718255	

### 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	28	64	100	161	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 even half of the amounts given can cause drastic overload, in other cases the maximum amounts can be even higher still giving acceptable separations.

# Packed columns

## NUCLEODUR® C<sub>18</sub> ec

Pore size 110 Å; octadecyl phase, endcapped, 17.5% C;  
eluent in column acetonitrile - water

Length →	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® 100-3 C<sub>18</sub> ec</b>						particle size 3 µm
EC analytical columns						
2 mm ID	760050.20		760054.20	760051.20	760053.20	760052.20
3 mm ID	760050.30		760054.30	760051.30	760053.30	760052.30
4 mm ID	760050.40		760054.40	760051.40	760053.40	760052.40
4.6 mm ID	760050.46	760046.46	760054.46	760051.46	760053.46	760052.46
EC guard columns*		4 x 2 mm: 761931.20		4 x 3 mm: 761931.30		
CC guard columns**		8 x 3 mm: 761005.30		8 x 4 mm: 761005.40		
<b>NUCLEODUR® 100-5 C<sub>18</sub> ec</b>						particle size 5 µm
Microbore analytical columns						
1 mm ID			717701.10	717700.10	717702.10	717703.10
EC analytical columns						
2 mm ID	760004.20		760013.20	760001.20	760008.20	760002.20
3 mm ID	760004.30		760013.30	760001.30	760008.30	760002.30
4 mm ID	760004.40		760013.40	760001.40	760008.40	760002.40
4.6 mm ID	760004.46	760035.46	760013.46	760001.46	760008.46	760002.46
EC guard columns*		4 x 2 mm: 761932.20		4 x 3 mm: 761932.30		
CC guard columns**		8 x 3 mm: 761100.30		8 x 4 mm: 761100.40		
VarioPrep preparative columns						
10 mm ID	762003.100			762029.100		762022.100
21 mm ID	762003.210			762029.210		762022.210
32 mm ID						762022.320
40 mm ID					762027.400	762022.400
VP guard columns***		10 x 8 mm: 762090.80		10 x 16 mm: 762090.160		
		15 x 32 mm: 762311.320		15 x 50 mm: 762311.500		
<b>NUCLEODUR® 100-10 C<sub>18</sub> ec</b>						particle size 10 µm
VarioPrep preparative columns						
10 mm ID	762011.100			762302.100		762010.100
21 mm ID	762011.210			762302.210		762010.210
32 mm ID						762010.320
40 mm ID					762303.400	762010.400
50 mm ID						762010.500
VP guard columns***		10 x 8 mm: 762090.80		10 x 16 mm: 762090.160		
		15 x 32 mm: 762311.320		15 x 50 mm: 762311.500		
Microbore, EC, and VarioPrep columns in packs of 1, guard columns see right						

## NUCLEODUR® C<sub>8</sub> ec

Pore size 110 Å; octyl phase, endcapped, 10.5% C;  
eluent in column acetonitrile - water

Length →	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® 100-3 C<sub>8</sub> ec</b>						particle size 3 µm
<b>EC analytical columns</b>						
2 mm ID	760063.20		760059.20	760060.20		760062.20
3 mm ID	760063.30		760059.30	760060.30		760062.30
4 mm ID	760063.40		760059.40	760060.40		760062.40
4.6 mm ID	760063.46	760064.46	760059.46	760060.46	760061.46	760062.46
EC guard columns*		4 x 2 mm: 761936.20		4 x 3 mm: 761936.30		
CC guard columns**		8 x 3 mm: 761012.30		8 x 4 mm: 761012.40		
<b>NUCLEODUR® 100-5 C<sub>8</sub> ec</b>						particle size 5 µm
<b>EC analytical columns</b>						
2 mm ID	760700.20		760704.20	760701.20		760703.20
3 mm ID	760700.30		760704.30	760701.30		760703.30
4 mm ID	760700.40		760704.40	760701.40		760703.40
4.6 mm ID	760700.46	760706.46	760704.46	760701.46	760702.46	760703.46
EC guard columns*		4 x 2 mm: 761937.20		4 x 3 mm: 761937.30		
CC guard columns**		8 x 3 mm: 761704.30		8 x 4 mm: 761704.40		
<b>VarioPrep preparative columns</b>						
10 mm ID	762072.100			762061.100		762062.100
21 mm ID	762072.210			762061.210		762062.210
32 mm ID						762062.320
40 mm ID					762079.400	762062.400
VP guard columns***	10 x 8 mm: 762092.80		10 x 16 mm: 762092.160		15 x 32 mm: 762321.320	
EC and VarioPrep columns in packs of 1, guard columns see below, Microbore columns with NUCLEODUR® C <sub>8</sub> ec on request!						

Guard column systems							Guard column holder
Guard columns for EC columns with ID			2 mm	3 mm	4 mm	4.6 mm	
*	Column Protection System (pack of)	EC	4/2 (3)	4/3 (3)	4/3 (3)	4/3 (3)	718966
**	ChromCart® guard columns (pack of)	CC	8/3 (3)	8/3 (3)	8/4 (3)	8/4 (3)	721359
Guard columns for VarioPrep columns with ID			8, 10 mm	16, 21 mm	32, 40 mm	≥ 50 mm	
***	VarioPrep guard columns (pack of)	VP	10/8 (2)	10/16 (2)	15/32 (1)	15/50 (1)	
	VP guard column holder		718251	718256	718253	718255	

# Packed columns

## NUCLEODUR® HILIC

Pore size 110 Å; zwitterionic phase for HILIC chromatography, 7% C; eluent in column acetonitrile - water 80:20

Length →	30 mm	50 mm	75 mm	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® HILIC, 1.8 µm</b>							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		760526.30			
4 mm ID	760521.40	760523.40		760526.40			
4.6 mm ID	760521.46	760523.46		760526.46			
EC guard columns*		4 x 2 mm: 761960.20		4 x 3 mm: 761960.30			
<b>NUCLEODUR® HILIC, 3 µm</b>							particle size 3 µm
EC columns							
2 mm ID		760532.20		760534.20	760531.20		760530.20
3 mm ID		760532.30		760534.30	760531.30		760530.30
4 mm ID		760532.40		760534.40	760531.40		760530.40
4.6 mm ID		760532.46		760534.46	760531.46	760533.46	760530.46
EC guard columns*		4 x 2 mm: 761961.20		4 x 3 mm: 761961.30			
CC guard columns**		8 x 3 mm: 761580.30		8 x 4 mm: 761580.40			
<b>NUCLEODUR® HILIC, 5 µm</b>							particle size 5 µm
EC columns							
2 mm ID		760552.20		760554.20	760551.20		760550.20
3 mm ID		760552.30		760554.30	760551.30		760550.30
4 mm ID		760552.40		760554.40	760551.40		760550.40
4.6 mm ID		760552.46		760554.46	760551.46	760553.46	760550.46
EC guard columns*		4 x 2 mm: 761962.20		4 x 3 mm: 761962.30			
CC guard columns**		8 x 3 mm: 761590.30		8 x 4 mm: 761590.40			
Columns in packs of 1, guard columns see right, Microbore columns and preparative columns with NUCLEODUR® HILIC on request!							

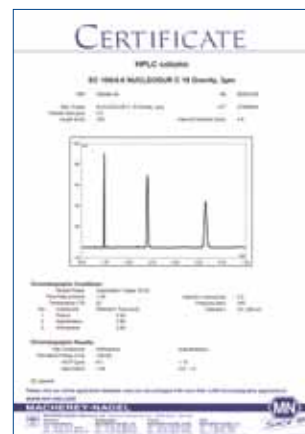
NUCLEODUR®

### 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® CN and CN-RP

Pore size 110 Å; cyano phase (nitrile), 7% C

Length →	50 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® 100-3 CN-RP</b> particle size 3 µm; eluent in column acetonitrile - water				
EC columns				
2 mm ID	760159.20	760157.20		
3 mm ID		760157.30		
4 mm ID			760156.40	
4.6 mm ID			760156.46	
EC guard columns*	4 x 2 mm: 761941.20		4 x 3 mm: 761941.30	
CC guard columns**	8 x 3 mm: 761430.30		8 x 4 mm: 761430.40	
<b>NUCLEODUR® 100-5 CN-RP</b> particle size 5 µm; eluent in column acetonitrile - water				
EC columns				
4 mm ID		760153.40		760152.40
4.6 mm ID		760153.46	760154.46	760152.46
EC guard columns*			4 x 3 mm: 761944.30	
CC guard columns**			8 x 4 mm: 761420.40	
<b>NUCLEODUR® 100-5 CN</b> particle size 5 µm; eluent in column <i>n</i> -heptane				
EC columns				
4 mm ID		760151.40	760149.40	760150.40
4.6 mm ID		760151.46	760149.46	760150.46
EC guard columns*			4 x 3 mm: 761943.30	
CC guard columns**			8 x 4 mm: 761419.40	

Columns in packs of 1, guard columns see below, Microbore columns and preparative columns with NUCLEODUR® CN / CN-RP on request!

Guard column systems						Guard column holder	
Guard columns for EC columns with ID		2 mm	3 mm	4 mm	4.6 mm		
*	Column Protection System (pack of)	EC	4/2 (3)	4/3 (3)	4/3 (3)	4/3 (3)	718966
**	ChromCart® guard columns (pack of)	CC	8/3 (3)	8/3 (3)	8/4 (3)	8/4 (3)	721359
Guard columns for VarioPrep columns with ID		8, 10 mm	16, 21 mm	32, 40 mm	≥ 50 mm		
***	VarioPrep guard columns (pack of)	VP	10/8 (2)	10/16 (2)	15/32 (1)	15/50 (1)	
	VP guard column holder		718251	718256	718253	718255	

# Packed columns

## NUCLEODUR® NH<sub>2</sub> and NH<sub>2</sub>-RP

Pore size 110 Å; amino phase, 2.5% C

Length →	100 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® 100-3 NH<sub>2</sub>-RP</b> particle size 3 µm; eluent in column acetonitrile - water				
EC columns				
2 mm ID	760740.20	760741.20		
4.6 mm ID			760742.46	760739.46
EC guard columns*	4 x 2 mm: 761951.20		4 x 3 mm: 761951.30	
CC guard columns**	8 x 3 mm: 761035.30		8 x 4 mm: 761035.40	
<b>NUCLEODUR® 100-5 NH<sub>2</sub>-RP</b> particle size 5 µm; eluent in column acetonitrile - water				
EC columns				
2 mm ID		760730.20		760732.20
3 mm ID		760730.30		760732.30
4 mm ID		760730.40		760732.40
4.6 mm ID		760730.46	760731.46	760732.46
EC guard columns*	4 x 2 mm: 761953.20		4 x 3 mm: 761953.30	
CC guard columns**	8 x 3 mm: 761137.30		8 x 4 mm: 761137.40	
<b>NUCLEODUR® 100-5 NH<sub>2</sub></b> particle size 5 µm; eluent in column <i>n</i> -heptane				
EC columns				
4 mm ID		760720.40		760722.40
4.6 mm ID		760720.46	760721.46	760722.46
EC guard columns*			4 x 3 mm: 761952.30	
CC guard columns**			8 x 4 mm: 761130.40	

Columns in packs of 1, guard columns see page 83, Microbore and preparative columns with NUCLEODUR® NH<sub>2</sub> / NH<sub>2</sub>-RP on request!

## Unmodified NUCLEODUR®

Pore size 110 Å; unmodified;  
eluent in column *n*-heptane

Length →	50 mm	125 mm	150 mm	250 mm
<b>NUCLEODUR® 100-3</b> particle size 3 µm				
EC analytical columns				
4.6 mm ID	760170.46		760172.46	760173.46
EC guard columns*			4 x 3 mm: 761966.30	
CC guard columns**			8 x 4 mm: 761007.40	
<b>NUCLEODUR® 100-5</b> particle size 5 µm				
EC analytical columns				
4 mm ID				760007.40
4.6 mm ID	760023.46		760012.46	760007.46
EC guard columns*			4 x 3 mm: 761967.30	
CC guard columns**			8 x 4 mm: 761055.40	
VarioPrep preparative columns				
10 mm ID	762077.100	762078.100		762007.100
21 mm ID	762077.210	762078.210		762007.210
40 mm ID			762075.400	762007.400
VP guard columns***	10 x 8 mm: 762094.80		10 x 16 mm: 762094.160	15 x 32 mm: 762330.320

Columns in packs of 1, guard columns see page 83, Microbore columns with unmodified NUCLEODUR® on request!

## EC standard columns for analytical HPLC

### Features

- 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 adapter  
Column heads SW 12, with inner threads M8 x 0.75 and UNF 10-32 (= 1/16" fitting)
- As screw-on guard column system we recommend the **Column Protection System** used with EC guard column cartridges with 4 mm length (see page 86).
- As built-in guard columns ChromCart® guard column cartridges with 8 mm length can be used with the guard column adapter EC (see page 87).
- Supplied with NUCLEODUR®, NUCLEOSHELL and NUCLEOSIL® spherical silicas



### Available standard dimensions of EC columns

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

Please ask for availability of certain phases

### Guard columns for EC columns

(pack of 3 each)		EC columns with ID				Use guard column holder
		2 mm	3 mm	4 mm	4.6 mm	
EC guard columns for Column Protection System guard column holder	EC	4/2	4/3	4/3	4/3	REF 718966
ChromCart® guard columns for EC guard column holder	CC	8/3	8/3	8/4	8/4	REF 721359

### Guard column systems, accessories and replacement parts for EC columns • Ordering information

Description	Pack of	REF	
Column Protection System	1 kit	718966	see figures and description on page 86
Guard column adapter EC	1	721359	see figures and description on page 87
EC fitting adapter	1	718987	
EC column head (nut)	1	718988	
EC PTFE sealing ring	4	718992	
3-part sealing combination for EC columns	5 kits	718998	

# Packed columns

## Column Protection System

Innovative and universal screw-on guard column holder system  
Suitable for all analytical HPLC columns with 1/16" fittings

### Features

- Cartridges filled with specified NUCLEODUR®, NUCLEOSIL® and NUCLEOSHELL HPLC adsorbents
- Ideal protection for your analytical main column → significant increase in column lifetime
- Minimized void volume → suitable also for ultra fast HPLC
- Special ferrules → pressure stability up to 1034 bar (15 000 psi)
- Visual contamination check → in-time changing of the guard column
- Guard column length 4 mm, ID 2 mm (for main columns with 2 mm ID) or ID 3 mm (for main columns with 3, 4 and 4.6 mm ID)
- UNIVERSAL RP guard columns suitable for all HPLC columns under RP conditions



### Content of the Column Protection System



Description	REF
<b>Column Protection System</b>	<b>718966</b>
Details	Content
Cartridge Holder	1
Capillaries	2
Ferrules	3
Wrenches	2
Manual	1

### Replacement parts for the Column Protection System • Ordering information

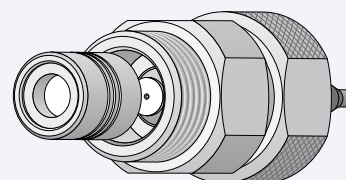
Description	Pack of	REF
Ferrules	5	718967
Replacement connector including O-ring	1	718968
Capillary tubes, nuts and metal ferrules	3	718969
Wrench (size 12 and 14 mm)	1	718970
EC 4/2 UNIVERSAL RP guard column (for main columns with 2 mm ID)	3	728777.20
EC 4/3 UNIVERSAL RP guard column (for main columns with 3, 4 and 4.6 mm ID)	3	728777.30

### Visual Contamination Check

**The cartridge is fitted with a white filter membrane:**

A discoloration of the filter membrane usually indicates that the cartridge should be replaced.

In case of colorless contaminants, a rising back pressure and / or loss of chromatographic performance advise to change the guard column.



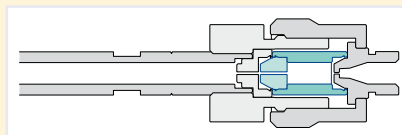


## EC guard column adapter

Standard guard column built-in adapter system  
Suitable for EC columns

### Features

- Cartridges filled with specified NUCLEODUR® and NUCLEOSIL® HPLC adsorbents
- Ideal protection for your analytical EC main column → significant increase of column lifetime
- Guard column length 8 mm, ID 3 mm (for main columns with 2 and 3 mm ID) or ID 4 mm (for main columns with 4 and 4.6 mm ID)



### EC guard column adapter · Ordering information

Description	Pack of	REF
EC guard column adapter	1	721359

### Installation of the EC guard column adapter



1. Unscrew the column head
2. Remove the fitting
3. Unscrew the EC guard column adapter



4. Screw the adapter sleeve onto the column
5. Insert the CC guard column
6. Screw the nut of the guard column adapter in place

# Packed columns

## VarioPrep (VP) columns for preparative HPLC

### Features

- Column system for preparative HPLC manufactured from stainless steel with two adjustable end fittings, e.g. 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 all NUCLEODUR® and NUCLEOSIL® spherical silicas

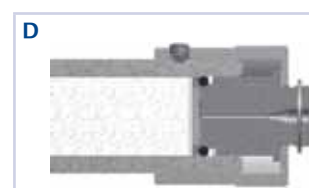
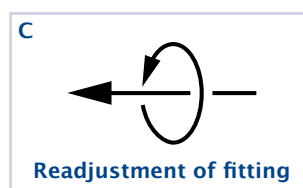
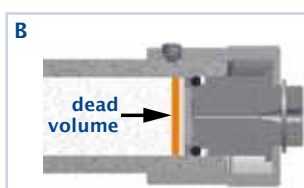
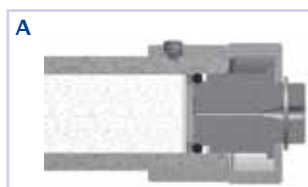


### Available standard dimensions of VarioPrep columns with axially adjustable end fittings

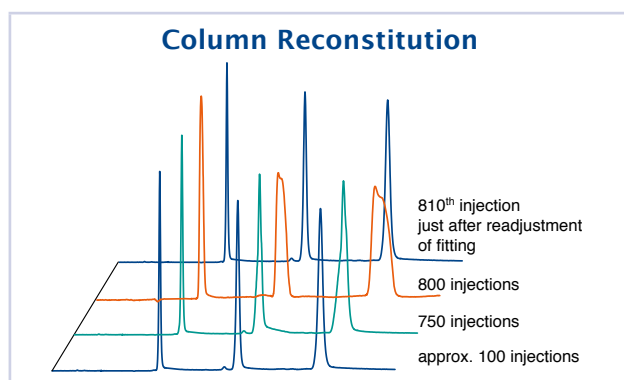
ID [mm]	Length [mm]		Length [mm]							End fitting design
	10*	15*	50	75	100	125	150	250	500	
8	+		+		+	+	+	+		
10			+		+	+	+	+		
16	+		+		+	+	+	+		
21			+	+	+	+	+	+		
32		+			+		+	+		
40			+		+	+	+	+	+	
50		+			+		+	+		
80								+	+	

\* 10 x 8, 10 x 16, 15 x 32 and 15 x 50 mm ID columns are used as guard columns and require adequate holders, see page 89.

### The VarioPrep principle

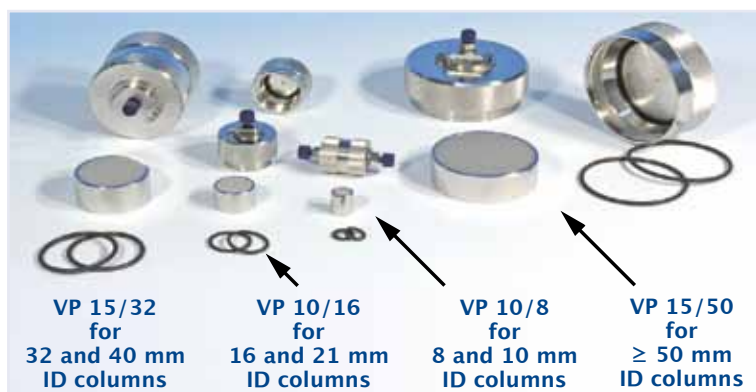


Based on our special packing procedure VarioPrep columns are produced with highest packing quality and bed density (A). Due to intensively chemical and/or mechanical exposure of the column adsorbent, shrinking of the column bed can occur (B; orange gap). In this even unlikely case readjustment of the VarioPrep column fitting (C; turning the nut at column inlet clockwise) will eliminate the emerged dead volume (D). The performance of the VarioPrep column is completely reconstituted and column lifetime is significantly extended.



### Reconstitution of VarioPrep column performance

- Slight peak broadening and deformation after 800 injections under strongly demanding conditions (pH 11; 50 °C; sample in DMSO)
- Readjustment of the column fitting restores the column performance and prolongs column lifetime noticeably

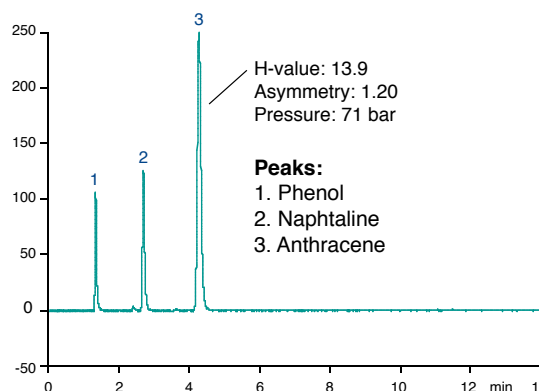
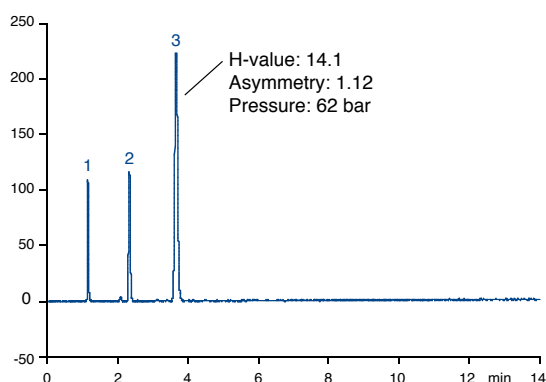


## The new guard column system for (semi-) preparative HPLC

- Easy handling and cartridge exchange
- Robust hardware
- Free rotary plunger fittings – low O-ring abrasion
- Cost-efficient cartridges
- Minimally invasive / no disturbance of the separation efficiency of main column
- Low back pressure
- Designed for pressures up to 400 bar

## Performance of preparative column when used with VarioPrep guard column

Columns: **125 x 16 mm NUCLEODUR® C<sub>18</sub> HTec, 5 μm**  
**125 x 16 mm NUCLEODUR® C<sub>18</sub> HTec, 5 μm + 10 x 16 mm NUCLEODUR® C<sub>18</sub> HTec guard column**  
 Eluent: acetonitrile – water (80:20, v/v)  
 Flow rate: 16 mL/min  
 Temperature: 22 °C



Using VarioPrep guard columns provides ideal protection of your main column – symmetry, pressure and retention stay almost constant.

## Technical data

- 1/16" thread
- free rotary plunger fittings – low O-ring abrasion
- stainless steel

Guard cartridge	Holder	Holder ID	Replacement O-ring (2 pcs)	Recommended for column ID	Preferred capillary ID	Typical flow rate
VP 10/8	REF 718251	8 mm	REF 718975	8 and 10 mm ID	0.17 and 0.25 mm	1–12 mL/min
VP 10/16	REF 718256	16 mm	REF 718976	16 and 21 mm ID	0.17, 0.25 and 0.5 mm	2–32 mL/min
VP 15/32	REF 718253	32 mm	REF 718977	32 and 40 mm ID	0.25, 0.5 and 1.0 mm	5–150 mL/min
VP 15/50	REF 718255	50 mm	REF 718978	≥ 50 mm ID	0.5 and 1.0 mm	20–250 mL/min

## Guard columns for VarioPrep columns • Ordering information

VP guard columns for VarioPrep guard column holder (pack of)	VP	VP columns with ID				Use guard column holder	Holder ID
		8, 10 mm	16, 21 mm	32, 40 mm	≥ 50 mm		
		10/8 (2)				REF 718251	8 mm
			10/16 (2)			REF 718256	16 mm
				15/32 (1)		REF 718253	32 mm
					15/50 (1)	REF 718255	50 mm



HPLC



GC



TLC



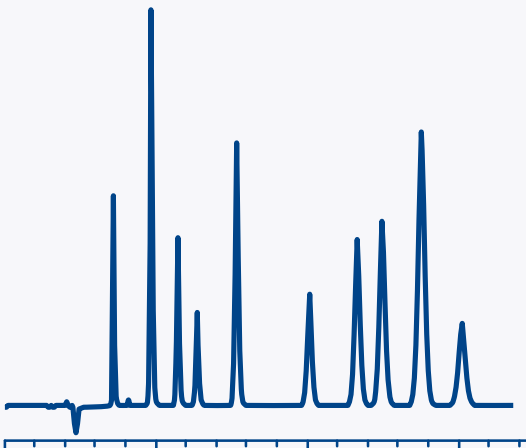
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