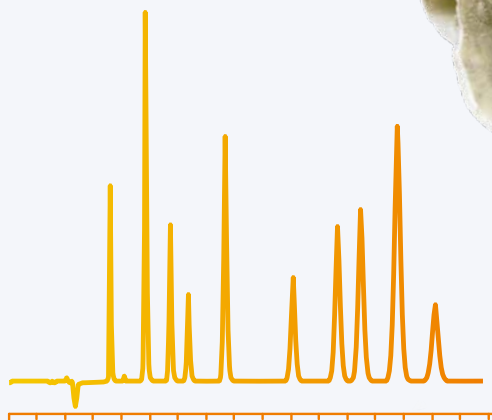


# Highest efficiency in HPLC by core-shell technology

NUCLEO SHELL®

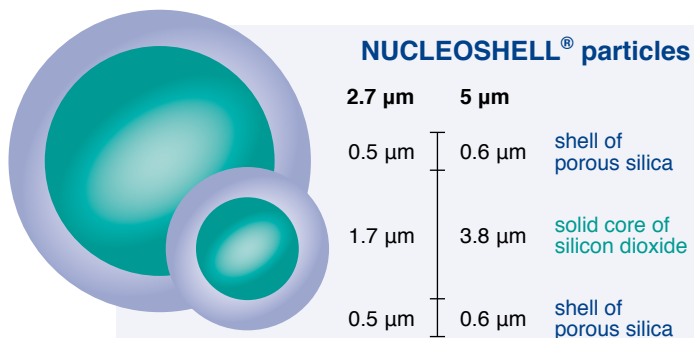


... we Meet your Needs



Improved  
Performance  
Enhanced  
Product range

# Ultrafast separations beyond high pressure driven UHPLC



## Core-shell technology

- Solid core of silicon dioxide, homogeneous shell of porous silica
- Highest efficiency compared to totally porous silica particles
- Particle size 2.7 µm (core 1.7 µm) and 5 µm (core 3.8 µm), pore size 90 Å, specific surface 130 (2.7 µm) and 90 (5 µm) m<sup>2</sup>/g; low back pressure enables use on conventional LC systems
- Pressure stability up to 600 bar

## NUCLEOSHELL® modifications

The program of NUCLEOSHELL® surface modifications now comprises the following phases:

- NUCLEOSHELL® RP 18
- NUCLEOSHELL® RP 18plus **NEW!**
- NUCLEOSHELL® Phenyl-Hexyl
- NUCLEOSHELL® PFP
- NUCLEOSHELL® HILIC

Several approaches have been made to achieve fast separations without losing chromatographic performance. HPLC columns packed with particles < 2 µm show very high efficiencies (plates/meter) and allow the use of smaller column sizes with the positive side effect of significant solvent saving. However they generate a high back pressure of the mobile phase during column runs which requires specifically designed equipment.

**NUCLEOSHELL® core-shell particle technology from MACHEREY-NAGEL is an alternate route to gain highest column efficiency and resolution at almost the same short run time but with much lower back pressure.**

NUCLEOSHELL® silica particles consist of a non-porous solid core and a porous outer shell – it is available in two particles sizes, 2.7 and 5 µm, respectively.

With conventional fully porous particles the mass transfer between stationary and mobile phase usually results

in peak broadening at higher flow rates (C-term in van Deemter equation). The short diffusion paths in the core-shell particles reduce the dwell time of the analyte molecules in the stationary phase, so that even at high flow velocities of the mobile phase, optimal separation results can be obtained. The van Deemter plots on page 3 demonstrate how efficiency is affected by flow rate. In comparison with fully porous silicas, core-shell particles from various manufacturers maintain the efficiency optimum (max. plates/m) over a long range of increasing linear mobile phase velocity.

## Theoretical column efficiency (optimal conditions)

Silica phase	d <sub>p</sub> [µm]	L [m]	HETP [µm]	Efficiency [plates/m]	L [mm]	N	R <sub>s</sub>	Analysis time
<b>NUCLEOSHELL®</b>	<b>2.7</b>	<b>1</b>	<b>4</b>	<b>250 000</b>	<b>100</b>	<b>25 000</b>	<b>112 %</b>	<b>40 %</b>
	<b>5</b>	<b>1</b>	<b>6.5</b>	<b>154 000</b>	<b>150</b>	<b>23 000</b>	<b>115 %</b>	<b>60 %</b>
NUCLEODUR®	1.8	1	4.5	222 222	100	22 000	105 %	40 %
	3	1	7.5	133 333	150	20 000	100 %	60 %
	5	1	12.5	80 000	250	20 000	100 %	100 %

## Benefits of core-shell technology

### Short diffusion paths

- Fast mass transfer (term C of Van Deemter equation)
- High flow velocity without peak broadening for fast LC

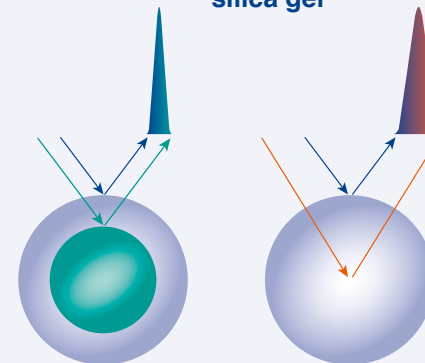
### Narrow particle size distribution (d<sub>90</sub>/d<sub>10</sub> ~ 1.1)

- Stable packing

### High heat transfer

- Minimized influence of frictional heat
- Efficiency of NUCLEOSHELL® ~ 250 000 m<sup>-1</sup> (HETP ~ 4 µm)

## Core-shell particles vs. totally porous silica gel



# Core-shell silica

Demands on HPLC separations are constantly increasing with respect to separation efficiency, detection limits, and the time requirements for each analysis. Core-shell technology sets new standards for analyses in research and quality control.

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

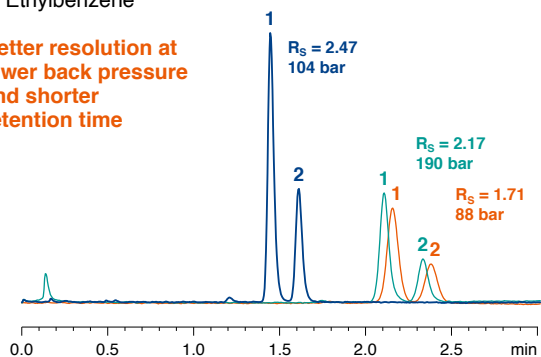
$R_s$  = Resolution  
 $\alpha$  = Selectivity  
 $k_i'$  = Retention  
 $N$  = Theoretical plates  $N \propto 1/d_p$   
 $d_p$  = Particle size

## Resolution $R_s$ as function of particle size

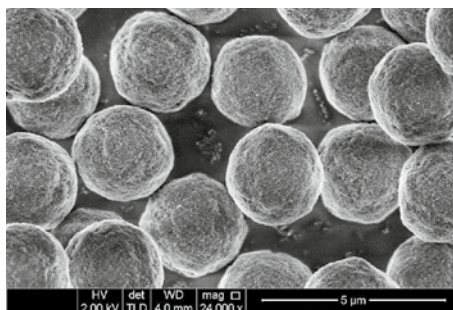
Columns: 50 x 4 mm each  
**NUCLEOSHELL® RP 18, 2.7  $\mu$ m**  
**NUCLEODUR® C<sub>18</sub> Gravity, 3  $\mu$ m**  
**NUCLEODUR® C<sub>18</sub> Gravity, 1.8  $\mu$ m**  
 Eluent: acetonitrile – water (60:40, v/v)  
 Flow rate: 1 mL/min  
 Temperature: 25 °C  
 Detection: UV, 254 nm

**Peaks:**  
 1. Naphthalene  
 2. Ethylbenzene

**Better resolution at lower back pressure and shorter retention time**



MN Appl. No. 125270

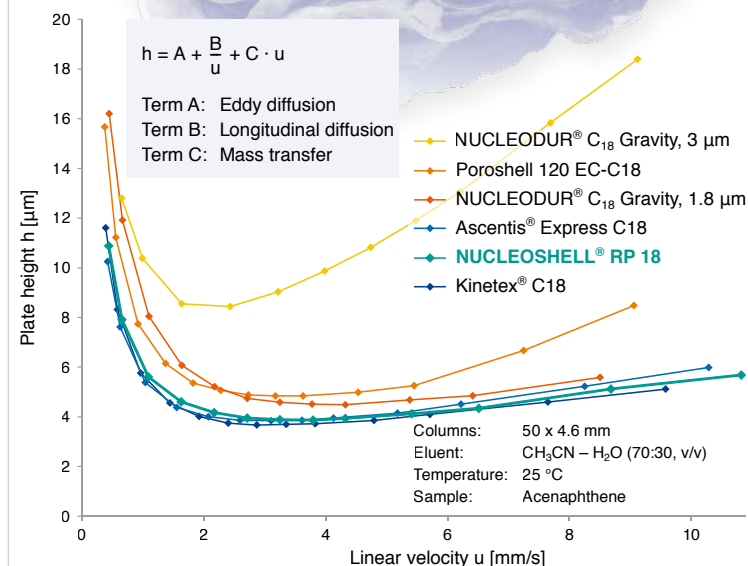


## Electron microscopic image of NUCLEOSHELL® particles

Utilizing a proprietary process of synthesis, NUCLEOSHELL® particles exhibit a distinct narrow particle size distribution ( $d_{90}/d_{10} \sim 1.1$ ). Columns packed with NUCLEOSHELL® core shell particles feature exceptional separation efficiencies with theoretical plate numbers easily comparable to totally porous sub 2 micron particles.

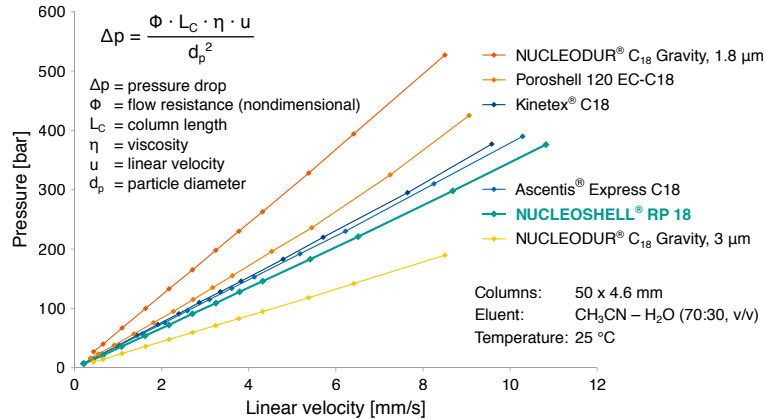
# NUCLEOSHELL®

## Van Deemter plots



MN Appl. No. 125500

## Pressure drop



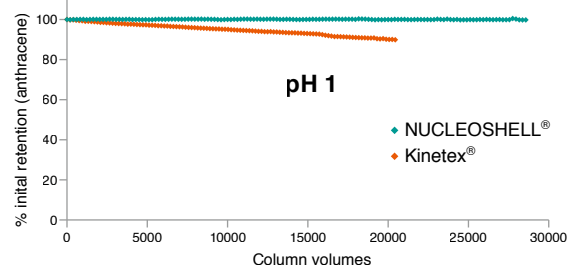
MN Appl. No. 125510

In direct comparison with the “conventional” sub 2 micron phases, NUCLEOSHELL® columns only generate about 60 % of the back pressure and can be operated with the majority of conventional HPLC systems. In order to develop the maximum performance of NUCLEOSHELL® columns, we recommend reducing extra column voids by using suitable capillaries (<0.15 mm inner diameter) and specially adapted detector cells. Moreover detector settings should be optimized by increasing the measuring rate or by decrease of the time constant.

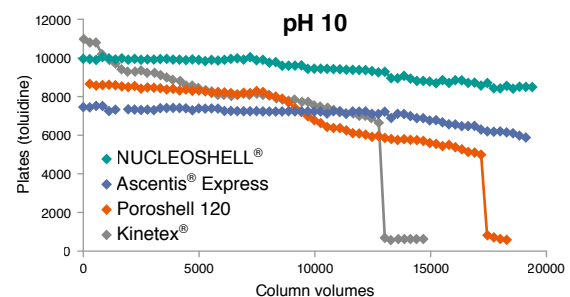
# Features of core-shell silica particles

## Stability under acidic and basic conditions

Column: 50 x 4.6 mm NUCLEOSHELL® RP 18, 2.7 µm  
 50 x 4.6 mm Kinetex® 2.6 µm C18  
 Eluent: acetonitrile – 1% TFA in H<sub>2</sub>O, pH 1 (50:50, v/v)  
 Flow rate: 1.3 mL/min; temperature 80 °C  
 Detection: UV, 254 nm  
 Sample: anthracene



Columns: 50 x 4.6 mm NUCLEOSHELL® RP 18, 2.7 µm  
 50 x 4.6 mm Ascentis® Express C18, 2.7 µm  
 50 x 4.6 mm Poroshell 120 EC-C18  
 50 x 4.6 mm Kinetex® 2.6 µm C18  
 Eluent: 20 mmol/L Na borate – 10 mmol/L NaOH – methanol, pH 10 (21:49:30, v/v)  
 Flow rate: 1.5 mL/min; temperature 40 °C  
 Detection: UV, 220 nm  
 Sample: toluidine



MN Appl. No. 125520/125530

The above figure shows a column stability test of NUCLEOSHELL® RP 18 at mobile phase levels pH 1 and pH 10 compared with three competing phases.

A criterion for the long-term stability of the column at pH extremes is the percentage decrease of initial retention and initial plates, respectively.

The column can also be operated at elevated temperatures without loss in retention behavior, efficiency or peak symmetry.

## Temperature stability

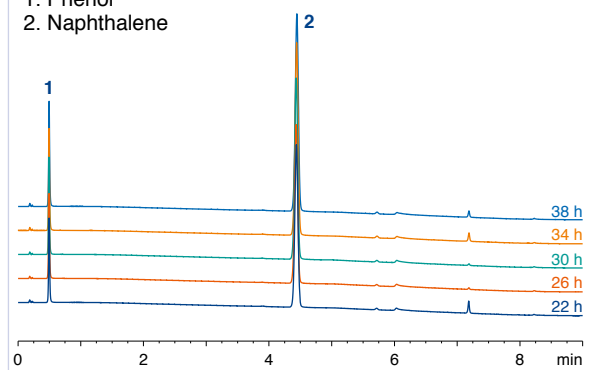
### Stability test:

Column: 50 x 2 mm NUCLEOSHELL® RP 18, 2.7 µm  
 Eluent: A) 10 mmol/L ammonium formate – methanol (9:1, v/v) + 120 µL formic acid, ~ pH 4  
 B) 10 mmol/L ammonium formate – methanol (1:9, v/v) + 120 µL formic acid, ~ pH 4  
 0–100% B in 7 min

Flow rate: 0.5 mL/min  
 Temperature: 100 °C  
 Detection: UV, 220 nm

### Peaks:

1. Phenol
2. Naphthalene



MN Appl. No. 125400

### Efficiency test:

Eluent: acetonitrile – water (60:40, v/v)  
 Flow rate: 0.33 mL/min  
 Temperature: 25 °C  
 Detection: UV, 254 nm  
 Sample: anthracene

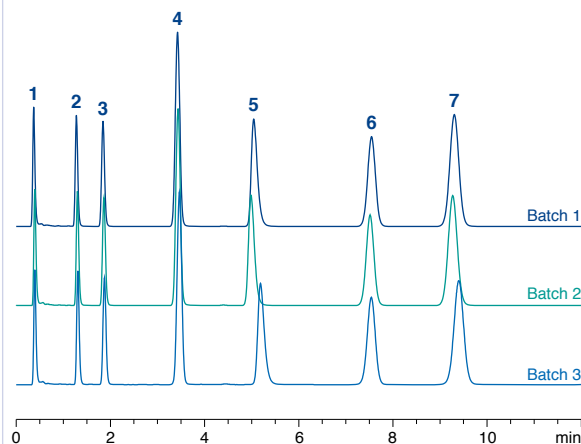
	HETP [µm]	Asymmetry
Start (t = 0)	5.2	0.98
End (t = 40 h)	5.2	1.01

## Batch-to-batch reproducibility

Column: 50 x 4 mm NUCLEOSHELL® RP 18, 2.7 µm  
 Eluent: methanol – 25 mmol/L KH<sub>2</sub>PO<sub>4</sub> pH 7 (70:30, v/v)  
 Flow rate: 1 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm

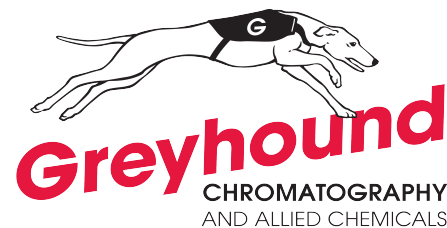
### Peaks:

1. Uracil
2. Toluene
3. Ethylbenzene
4. Acenaphthene
5. Amitriptyline
6. o-Terphenyl
7. Triphenylene



MN Appl. No. 125410

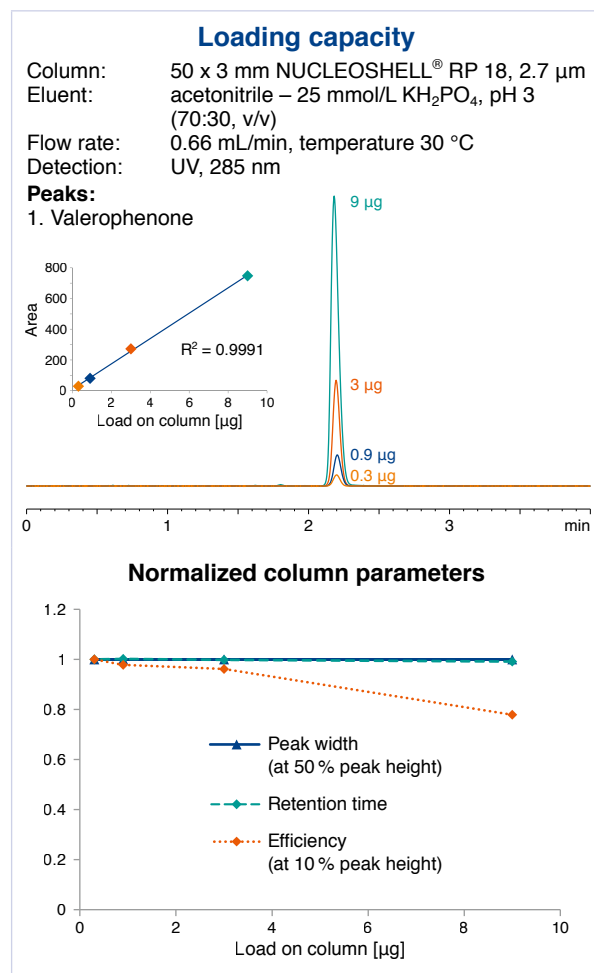
Uniformly shaped NUCLEOSHELL® particles combined with optimized bonding technology safeguard tightly packed columns for 100% reproducible results.





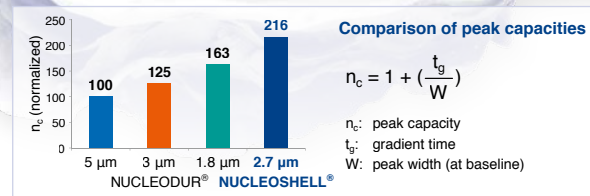
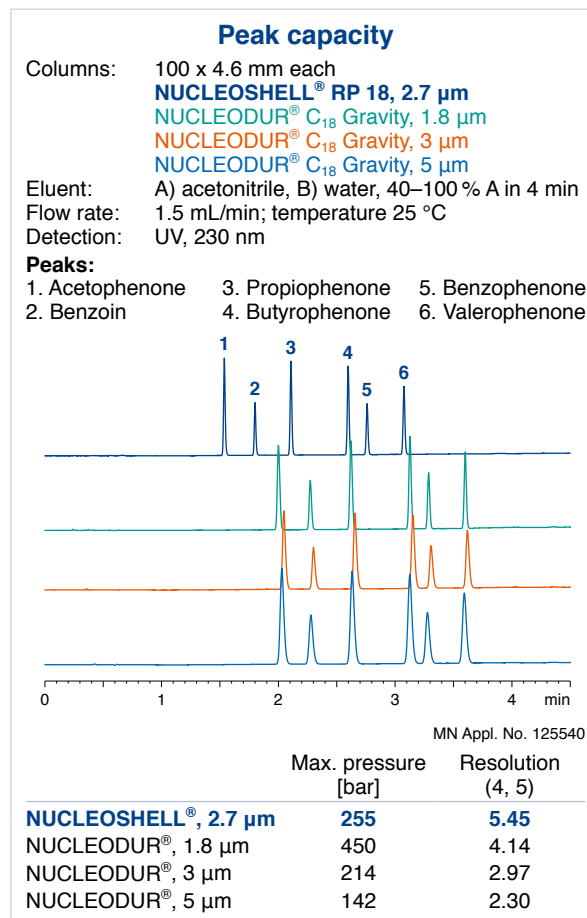
## Loading capacity

NUCLEOSHELL® columns allow **reliable quantification** in a wide analytical detection range. Retention time and peak width at 50% height remain constant with increasing column load although core-shell particles are suspected of showing a slightly lower loading capacity compared to fully porous silica materials.



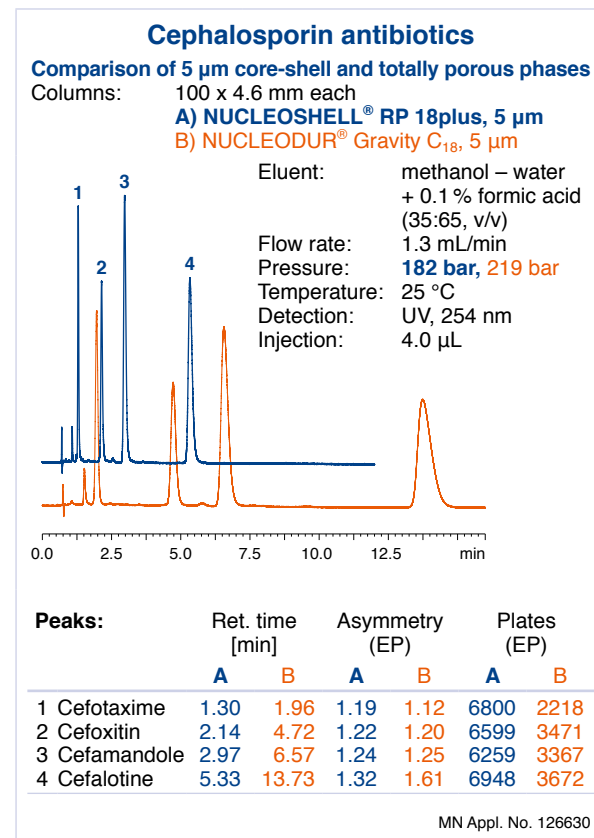
## Peak capacity

The peak capacity is a measure of the number of sample analytes that can be separated on HPLC columns per time unit. Narrow peaks increase the peak capacity and efficiency of analytical columns. The example shows, that in comparison with totally porous NUCLEODUR® silica (1.8 µm) NUCLEOSHELL® provides 33% higher peak capacity.



## Method transfer of 5 µm particle columns

NUCLEOSHELL® is also available in 5 µm particle size to offer all benefits of core-shell technology to all applications which are bound to particle size.



# NUCLEOSHELL® RP 18

## Key features:

- Based on core-shell particle technology for fast and efficient HPLC
- Suitable for LC/MS and HPLC at pH extremes (pH 1–11)
- Superior base deactivation, ideal for method development

## Technical characteristics:

Octadecyl modification, multi-endcapped; pore size 90 Å, available particle sizes 2.7 µm and 5 µm, carbon content 7.8 % for 2.7 µm, 6.1 % for 5 µm

## Recommended application:

Overall sophisticated analytical separations, e.g., analgesics, anti-inflammatory drugs, antidepressants; herbicides; phytopharmaceuticals; immunosuppressants

USP L1

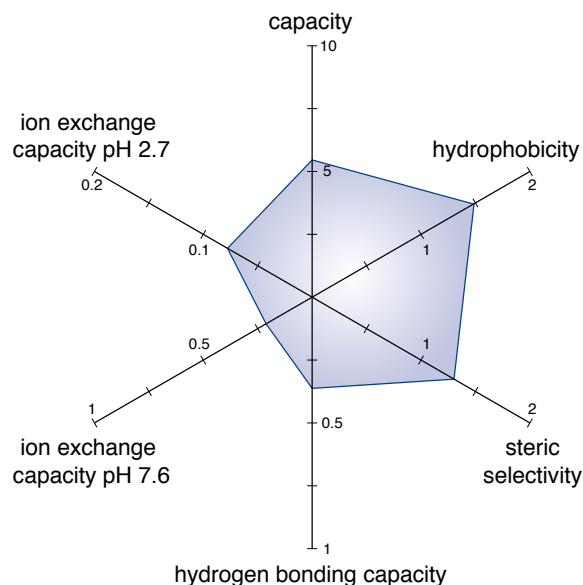
## NUCLEOSHELL® RP 18

NUCLEOSHELL® RP 18 is based on core-shell particle technology silica. A unique derivatization process generates a homogeneous surface with a high density of bonded silanes (carbon content 7.8 % for 2.7 µm, 6.1 % for 5 µm). The following thorough endcapping suppresses any unwanted polar interactions between the silica surface and the sample, which makes NUCLEOSHELL® RP 18 particularly suitable for the separation of basic and other ionizable analytes.

The extremely reduced silanol activity of the phase can be demonstrated by applying basic analytes, such as tricyclic antidepressants. The chromatogram on page 7 shows a sharp elution profile (superior resolution!) of these highly polar compounds with an excellent asymmetry value for amitriptyline of 1.12.

## Tanaka plot of NUCLEOSHELL® RP 18

The diagram below underlines the distinct hydrophobic characteristics and the low silanol activity of the stationary phase.



## Parameters of the Tanaka diagram

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 separation of 13  $\beta$ -lactam antibiotics illustrates how time of analysis can be shortened to a fractional part by using core-shell particles without loss of resolution at moderate back pressure.

## 13 $\beta$ -lactam antibiotics in less than 3 min

Columns: 50 x 4 mm NUCLEOSHELL® RP 18, 2.7 µm  
150 x 4 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm

Eluent: A) acetonitrile; B) 20 mmol/L KH<sub>2</sub>PO<sub>4</sub> pH 3.5  
10% A (0.5 min) → 50% A in 1.5 min (0.5 min 50% A)  
10% A (3 min) → 50% A in 9 min (3 min 50% A)

Flow rate: 2 mL/min, 1 mL/min

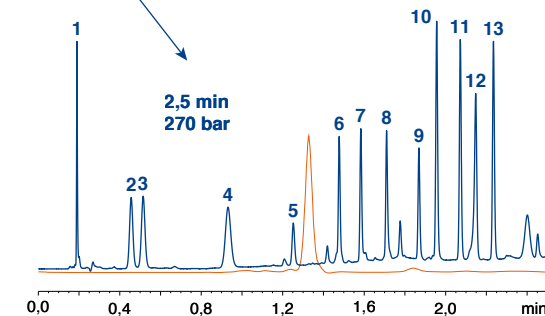
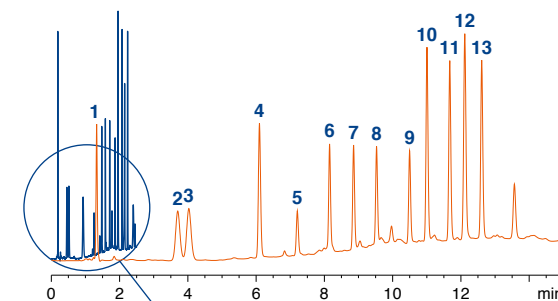
Pressure: 270 bar, 110 bar

Temperature: 25 °C

Detection: UV, 220 nm

## Peaks:

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



MN Appl. No. 124940

## Tricyclic antidepressants · comparison of selectivity and resolution

Columns: 50 x 4.6 mm each  
**NUCLEOSHELL® RP 18, 2.7 µm**  
 Ascentis® Express C18  
 Kinetex® 2.6 µm C18  
 Poroshell 120 EC-C18

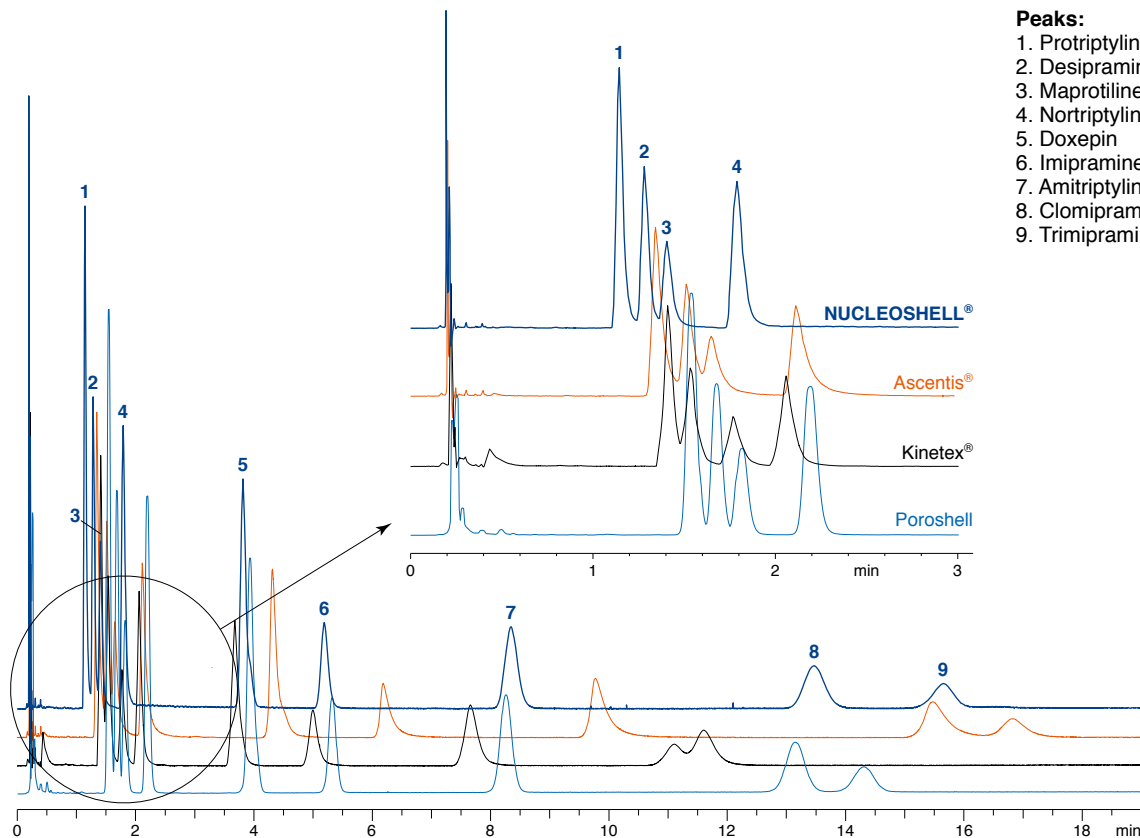
Eluent: methanol – acetonitrile – 25 mmol/L KH<sub>2</sub>PO<sub>4</sub> pH 7  
 (22.5:22.5:55, v/v/v)

Flow rate: 2 mL/min  
 Pressure: **224 bar, 239 bar, 248 bar, 212 bar**  
 Temperature: 40 °C  
 Detection: UV, 220 nm

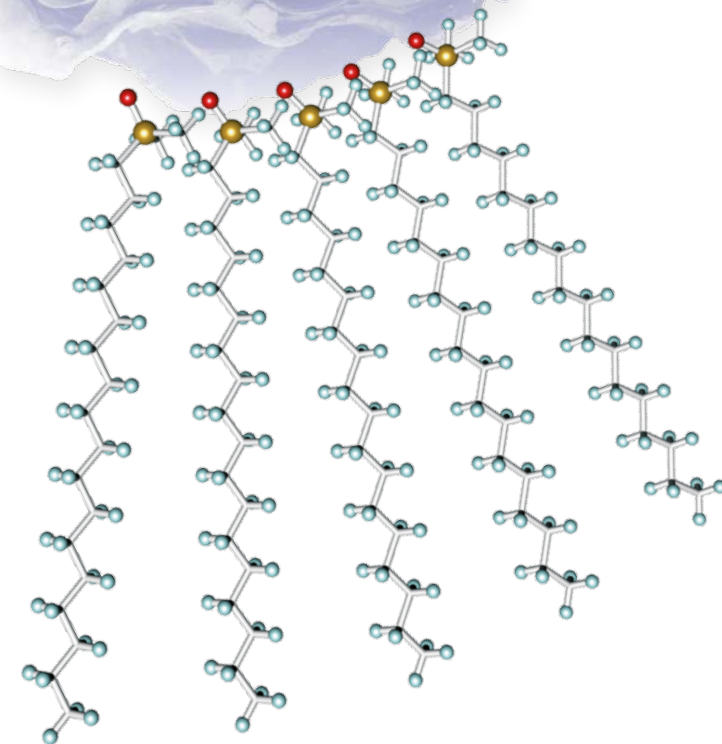
	Asymmetry (amitriptyline)	Resolution (8, 9)
<b>NUCLEOSHELL®</b>	<b>1.12</b>	<b>3.35</b>
Ascentis® Express	2.07	1.91
Kinetex®	1.33	n.a.
Poroshell	1.05	1.95

### Peaks:

1. Protriptyline
2. Desipramine
3. Maprotiline
4. Nortriptyline
5. Doxepin
6. Imipramine
7. Amitriptyline
8. Clomipramine
9. Trimipramine



MN Appl. No. 124960



NUCLEOSHELL® RP 18 combines innovative silica technology and excellent surface deactivation, that outperforms conventional C<sub>18</sub> silicas in terms of efficiency, resolution and speed. Due to the applied core-shell particle design the back pressure at elevated flow rates remains at a moderate level and permits the use of existing HPLC equipment in many cases.

**NUCLEOSHELL® RP 18 with extended pH stability, low bleed characteristics in LC/MS applications and overall robustness is an ideal tool for method development and routine analysis in modern HPLC.**

# NUCLEOSHELL® RP 18plus

## Key features:

- Based on core-shell particle technology for fast and efficient HPLC
- Hydrophobic C<sub>18</sub> phase with distinct polar selectivity, ideal for method development
- Excellent performance under highly aqueous conditions

## Technical characteristics:

Monomeric octadecyl modification, multi-encapped; pore size 90 Å, available particle sizes 2.7 µm and 5 µm, carbon content 5.7% for 2.7 µm, 4.4% for 5 µm; pH stability 2–9; suitable for LC/MS

## Recommended application:

Overall sophisticated analytical separations, especially for polar compounds, e.g., pharmaceuticals like antibiotics, water-soluble vitamins, organic acids

USP L1

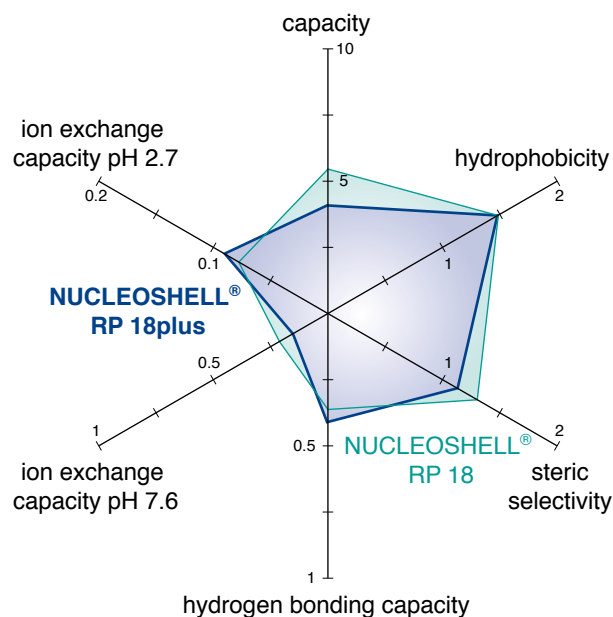
## NUCLEOSHELL® RP 18plus

NUCLEOSHELL® RP 18plus is a C<sub>18</sub> modified core-shell silica. Due to a monomeric bonding chemistry this HPLC phase offers hydrophobic characteristics with distinct polar selectivity. A special derivatization process generates a medium density of bonded silanes (carbon content 5.7% for 2.7 µm, 4.4% for 5 µm) with reduced steric selectivity compared to NUCLEOSHELL® RP 18. Due to its low bleeding characteristics NUCLEOSHELL® RP 18plus is suitable for LC/MS applications.

## Tanaka plot of NUCLEOSHELL® RP 18plus

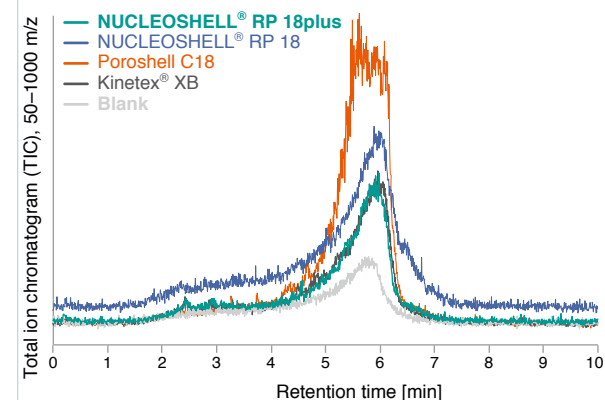
The more polar characteristics of NUCLEOSHELL® RP 18plus compared to NUCLEOSHELL RP 18 are demonstrated by the lower capacity (retention of pentylbenzene). Also a comparison of retention of the glycopeptide antibiotic vancomycin on several octadecyl modified core-shell phases (page 9) underlines the polar selectivity of NUCLEOSHELL® RP 18plus.

The extremely reduced silanol activity of the phase can be demonstrated by the low ion exchange capacity at pH 7.6 which indicates excellent endcapping (for parameters of the Tanaka plot see page 6). The radar diagram also shows that the monomeric bonding chemistry results in a thinner carbon coating and less steric selectivity.



## Bleeding characteristics

Column: 50 x 2 mm NUCLEOSHELL® RP 18plus, 2.7 µm  
Eluent: A) water + 0.1% formic acid, B) acetonitrile + 0.1% formic acid; 95% A → 5% A in 4.5 min (0.5 min) → 95% A in 0.5 min (4.5 min)  
Flow rate: 0.5 mL/min  
Temperature: 25 °C  
Detection: MS



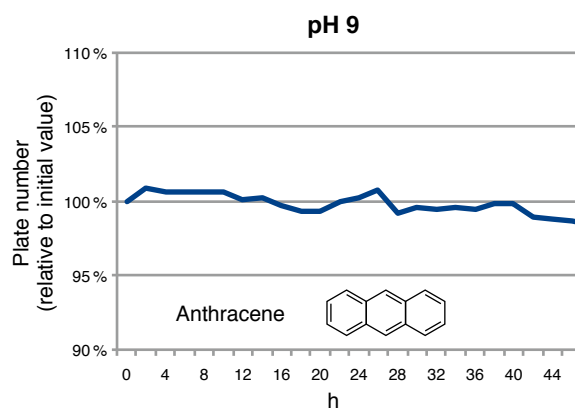
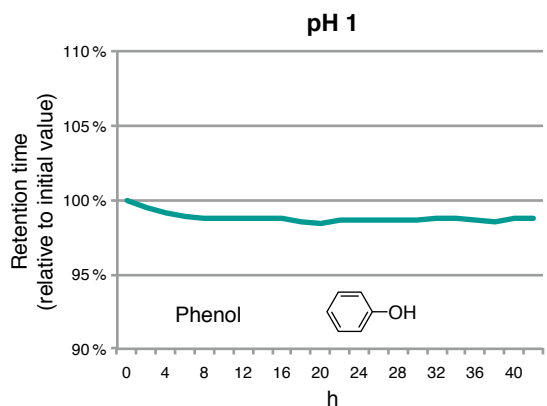
MN Appl. No. 126640

**NUCLEOSHELL® RP 18plus combines superbly hydrophobic and polar selectivity – so it is a useful tool for method development in RP chromatography. Good pH stability and low bleeding characteristics make it ideal especially for LC/MS applications.**



## pH stability of NUCLEOSHELL® RP 18plus

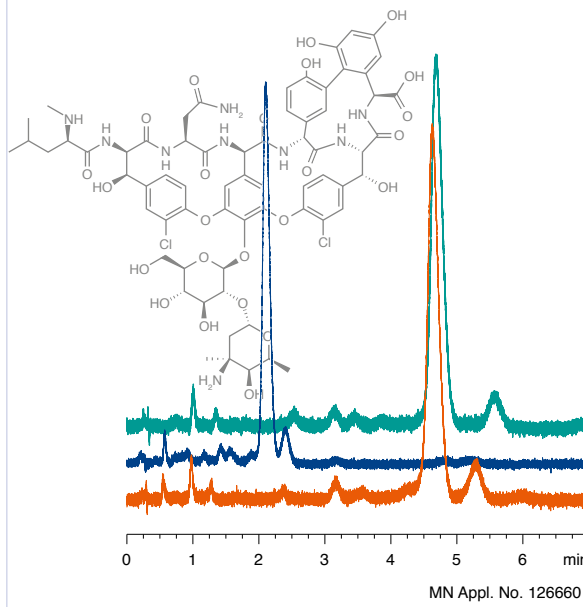
Column: 100 x 4 mm  
 NUCLEOSHELL® RP 18plus, 2.7 µm  
 Eluent **pH 1**: acetonitrile + 1% TFA – water + 1% TFA  
 pH 1 (50:50, v/v)  
 Eluent **pH 9**: 50 mmol/L triethylammonium acetate ad-  
 justed to pH 9  
 Flow rate: pH 1: 0.8 mL/min, pH 9: 0.56 mL/min  
 Temperature: pH 1: 60 °C, pH 9: 50 °C  
 Detection: UV, 254 nm  
 Injection: 1 µL



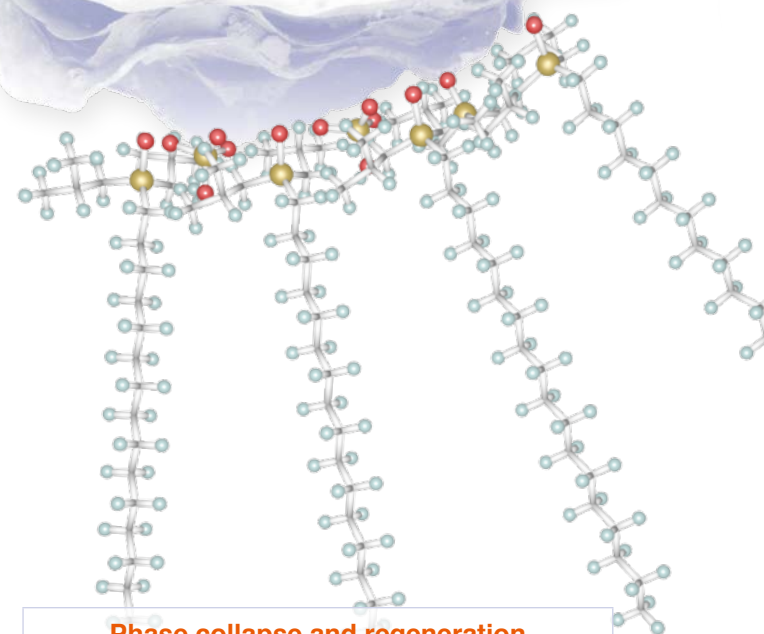
MN Appl. No. 126650

## Polar selectivity shown for vancomycin

Columns: 50 x 3 mm  
 NUCLEOSHELL® RP 18plus, 2.7 µm  
 NUCLEOSHELL® RP 18, 2.7 µm  
 Kinetex® 2.6 µm C18  
 Eluent: water – methanol – acetonitrile – glacial  
 acetic acid adjusted to pH 3.2 with NaOH  
 (100:8:2:0.3, v/v/v/v)  
 Flow rate: 0.9 mL/min  
 Temperature: 35 °C  
 Detection: UV, 240 nm  
 Injection: 10 µL, 10 µg/mL vancomycin

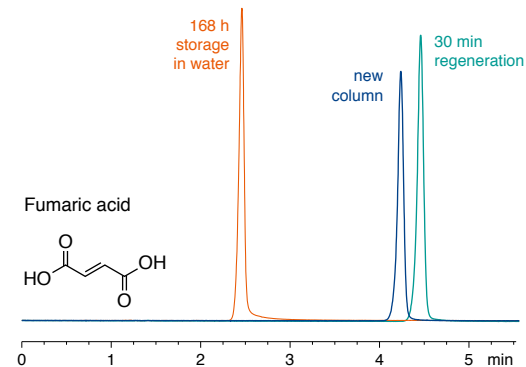


In addition NUCLEOSHELL® RP 18plus provides a good stability under highly aqueous conditions. Even by long term usage or storage of the phase hardly phase collapse and loss of retention are observed. The original performance can be regained after a short regeneration procedure.



## Phase collapse and regeneration

Column: 100 x 4 mm  
 NUCLEOSHELL® RP 18plus, 2.7 µm  
 Eluent: 20 mmol/L KH<sub>2</sub>PO<sub>4</sub> adjusted to pH 2.6  
 Flow rate: 0.5 mL/min, temperature 20 °C  
 Detection: UV, 215 nm, injection 0.5 µL



MN Appl. No. 126670

# NUCLEOSHELL® Phenyl-Hexyl

## Key features:

- Based on core-shell particle technology for fast and efficient HPLC
- Hydrophobic phase with alternative selectivity in comparison to classical C<sub>18</sub> modifications
- Separation principle based on 2 retention mechanisms:  $\pi$ - $\pi$  interactions and hydrophobic interactions

## Technical characteristics:

Phenyl-Hexyl modification, multi-encapped; pore size 90 Å, particle size 2.7 µm, carbon content 4.5%; pH stability 1–10; suitable for LC/MS

## Recommended application:

Aromatic and unsaturated compounds, polar compounds like pharmaceuticals, antibiotics

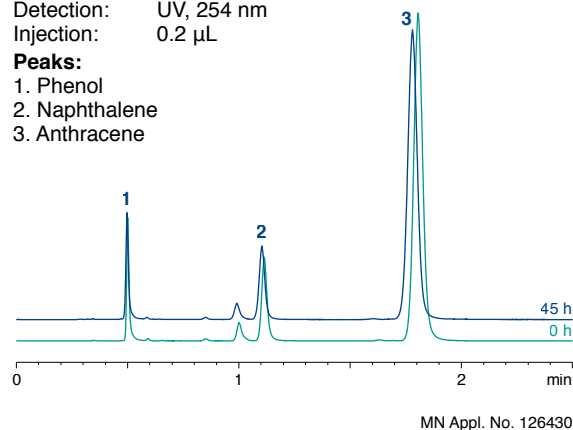
USP L11

Phenyl-Hexyl modified phases offer an excellent separation efficiency especially for aromatic and unsaturated compounds with electron-withdrawing groups. The combination of hydrophobic and  $\pi$ - $\pi$  interactions results in an alternative and interesting selectivity profile compared

## Temperature stability of NUCLEOSHELL® Phenyl-Hexyl

Column: 50 x 2 mm NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm  
 Eluent: acetonitrile – 10 mmol/L ammonium formate pH 4 (50:50, v/v)  
 Flow rate: 0.33 mL/min  
 Temperature: 100 °C  
 Detection: UV, 254 nm  
 Injection: 0.2 µL

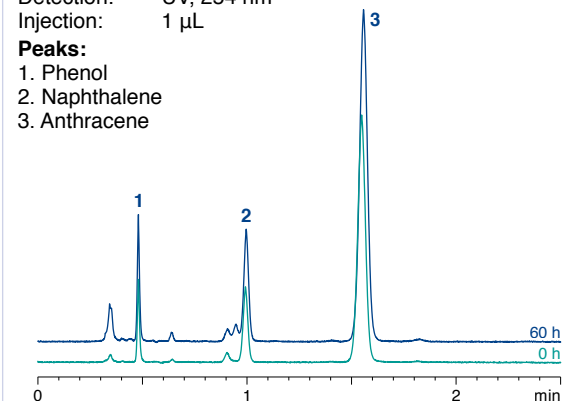
**Peaks:**  
 1. Phenol  
 2. Naphthalene  
 3. Anthracene



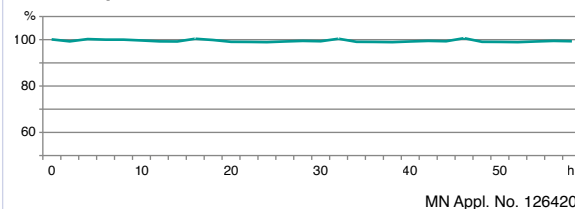
## Stability of NUCLEOSHELL® Phenyl-Hexyl at pH 10

Column: 50 x 4 mm NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm  
 Eluent: acetonitrile – 50 mmol/L TEA pH 10 (60:40, v/v); pH of the mixture 10.4  
 Flow rate: 1 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm  
 Injection: 1 µL

**Peaks:**  
 1. Phenol  
 2. Naphthalene  
 3. Anthracene



## Relative plate numbers

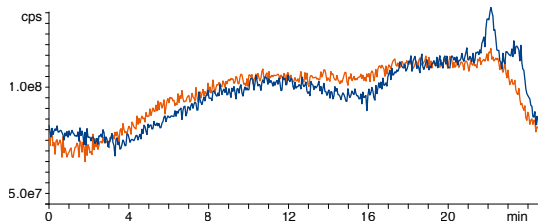


the C<sub>18</sub> or C<sub>8</sub> modifications. NUCLEOSHELL® Phenyl-Hexyl is based on an unique surface bonding chemistry - therefore it is suitable for LC/MS due to low bleeding characteristics and offers high temperature stability and pH stability from 1 to 10.

## Bleeding characteristics of NUCLEOSHELL® Phenyl-Hexyl

Columns: 50 x 2 mm each  
**NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm**  
 Kinetex® Phenyl-Hexyl  
 Eluent: A) acetonitrile, B) water;  
 5–95 % A in 25 min  
 Flow rate: 0.2 mL/min  
 Temperature: 25 °C  
 Detection: MS

MN Appl. No. 126400



NUCLEOSHELL® Phenyl-Hexyl is a robust phase with an alternative RP selectivity for aromatic and unsaturated analytes compared to classical C<sub>18</sub>/C<sub>8</sub> phases – it is an additional and useful tool for all chromatographers.

## Comparison of Phenyl-Hexyl phases for the separation of sulfonamides

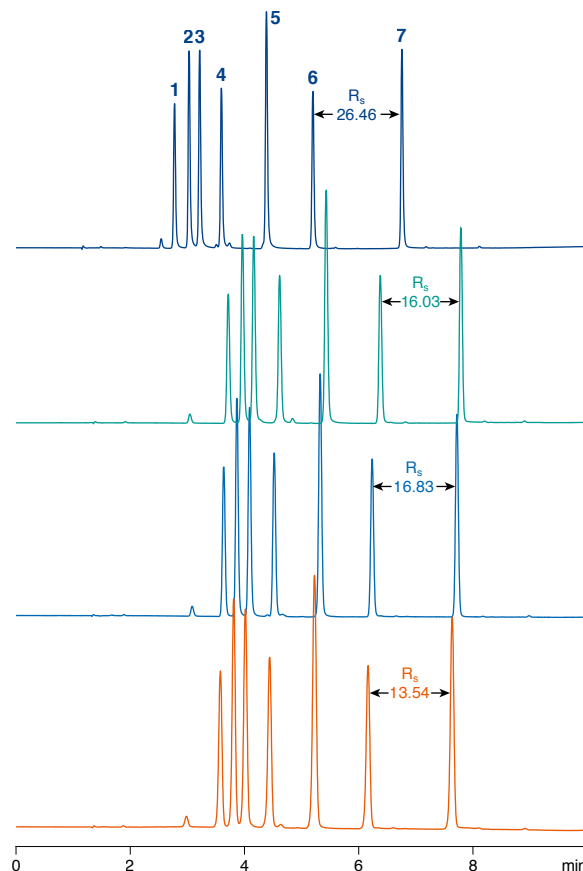
Columns: 150 x 3 mm each  
**NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm**  
 NUCLEODUR® Phenyl-Hexyl, 1.8 µm  
 NUCLEODUR® Phenyl-Hexyl, 3 µm  
 NUCLEODUR® Phenyl-Hexyl, 5 µm

Eluent: A) methanol,  
 B) 0.1 % formic acid in water,  
 20–80 % A in 10 min

Flow rate: 0.56 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm  
 Injection: 0.5 µL

**Peaks:**  
 1. Sulfadiazine  
 2. Sulfachloropyridazine  
 3. Sulfapyridine  
 4. Sulfamerazine  
 5. Sulfadimidine  
 6. Sulfathiazole  
 7. Sulfadimethoxine

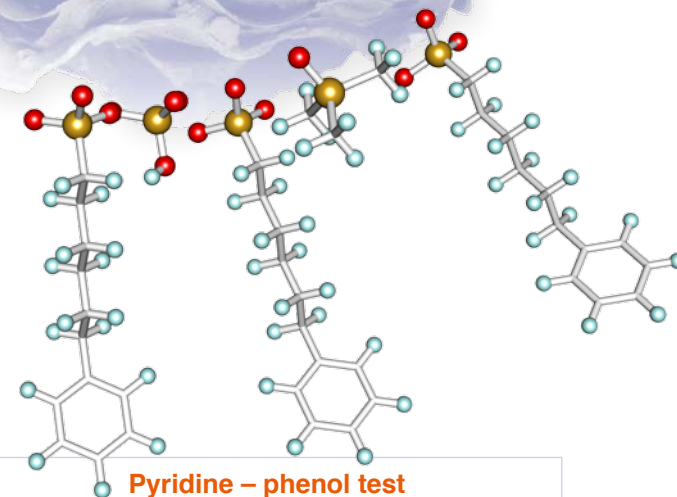
On NUCLEOSHELL® Phenyl-Hexyl the resolution of the last two peaks is higher than on the fully porous 1.8 µm NUCLEODUR® Phenyl-Hexyl.



MN Appl. No. 125860

The separation of sulfonamides proves the scalability from fully porous NUCLEODUR® to NUCLEOSHELL® Phenyl-Hexyl. Hereby the core-shell silica exhibits under same conditions identical selectivity, narrower peaks and slightly shorter retention. Thus, method transferability between NUCLEODUR® and NUCLEOSHELL® is guaranteed, either for speeding up your methods or

scaling up for preparative requirements. The pyridine-phenol test shows that NUCLEOSHELL® Phenyl-Hexyl provides a symmetrical peak for pyridine and higher resolution in comparison to other core-shell based Phenyl-Hexyl phases, which underlines the excellent base deactivation.

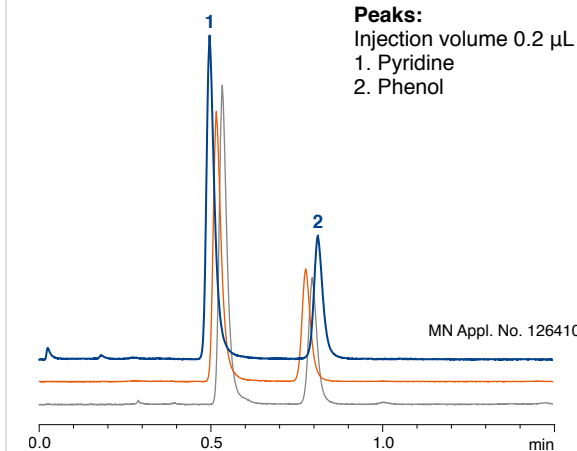


## Pyridine – phenol test

Columns: 50 x 2 mm each  
**NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm**  
 Kinetex® Phenyl-Hexyl  
 Ascentis® Express Phenyl-Hexyl

Eluent: acetonitrile – water (70:30, v/v)  
 Flow rate: 0.3 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm

**Peaks:**  
 Injection volume 0.2 µL  
 1. Pyridine  
 2. Phenol



MN Appl. No. 126410

## Key features:

- Based on core-shell particle technology for fast and efficient HPLC
- 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
  - $\pi$ - $\pi$  interactions
  - hydrophobic interactions

## Technical characteristics:

Pentafluorophenyl propyl modification, multi-encapped; pore size 90 Å, particle size 2.7 µm, carbon content ~ 3%; pH stability 1–9; suitable for LC/MS

## Recommended application:

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

USP L43

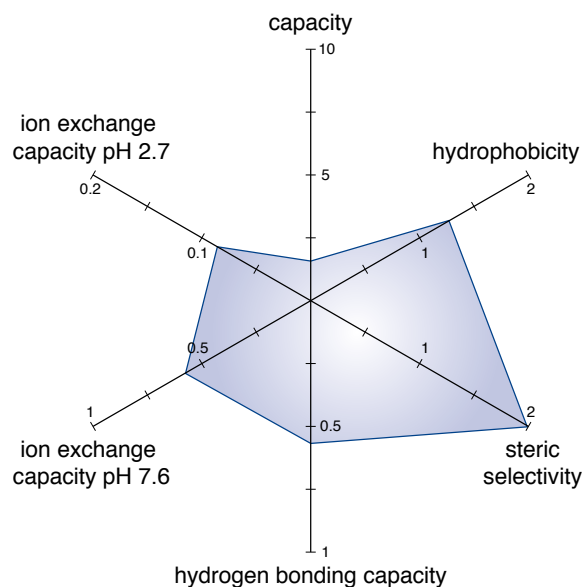
## Orthogonality in selectivity

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 F<sub>5</sub>). Especially the orthogonal selectivity compared to traditional alkyl phases widens the scope in analytical HPLC. Thus NUCLEOSHELL® PFP offers an excellent selectivity especially for highly polar analytes, 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 NUCLEOSHELL® PFP offers four different retention mechanisms: polar interactions (H bonds), dipole-dipole interactions,  $\pi$ - $\pi$  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 NUCLEOSHELL® PFP



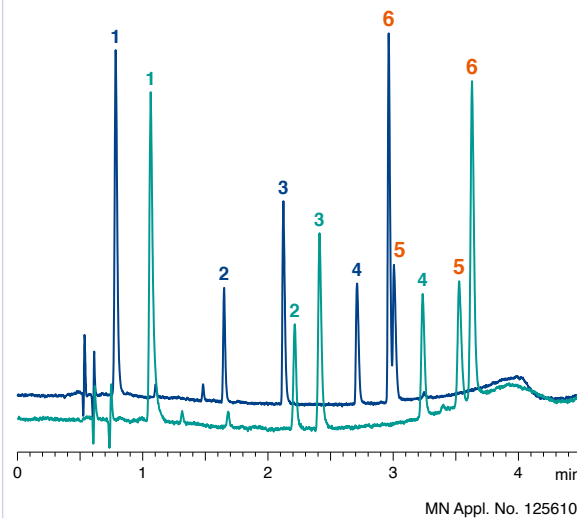
## β-Blockers · orthogonal selectivity of NUCLEOSHELL® PFP

Columns: 100 x 4.6 mm each  
 NUCLEOSHELL® RP 18, 2.7 µm  
 NUCLEOSHELL® PFP, 2.7 µm

Eluents: A) acetonitrile + 0.1 % formic acid;  
 B) 0.1 % formic acid;  
 10–35 % A in 2.5 min, 35–50 % A in 2 min

Flow rate: 1.7 mL/min  
 Temperature: 25 °C  
 Detection: UV, 280 nm

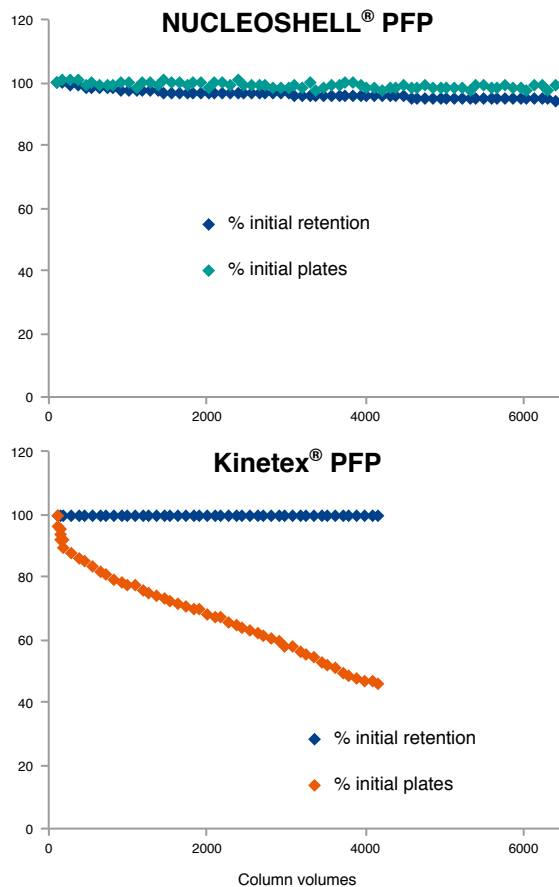
- Peaks:
1. Atenolol
  2. Pindolol
  3. Metroprolol
  4. Labetalol
  5. Alprenolol
  6. Propranolol



NUCLEOSHELL® PFP combines the benefits of core-shell technology, high stability and orthogonal selectivity. So it is a useful complementary tool for highly efficient separations especially of isomers, halogenated, aromatic and/or polar compounds.

## Stability of NUCLEOSHELL® PFP at pH 1

Columns: **100 x 4.6 mm NUCLEOSHELL® PFP, 2.7 µm**  
**100 x 4.6 mm Kinetex® 2.6 µm PFP**  
 Eluent: acetonitrile – 0.5% TFA pH 1 (50:50, v/v)  
 Flow rate: 1.3 mL/min  
 Temperature: 60 °C  
 Detection: UV, 254 nm  
**Sample:**  
 Ethylbenzene

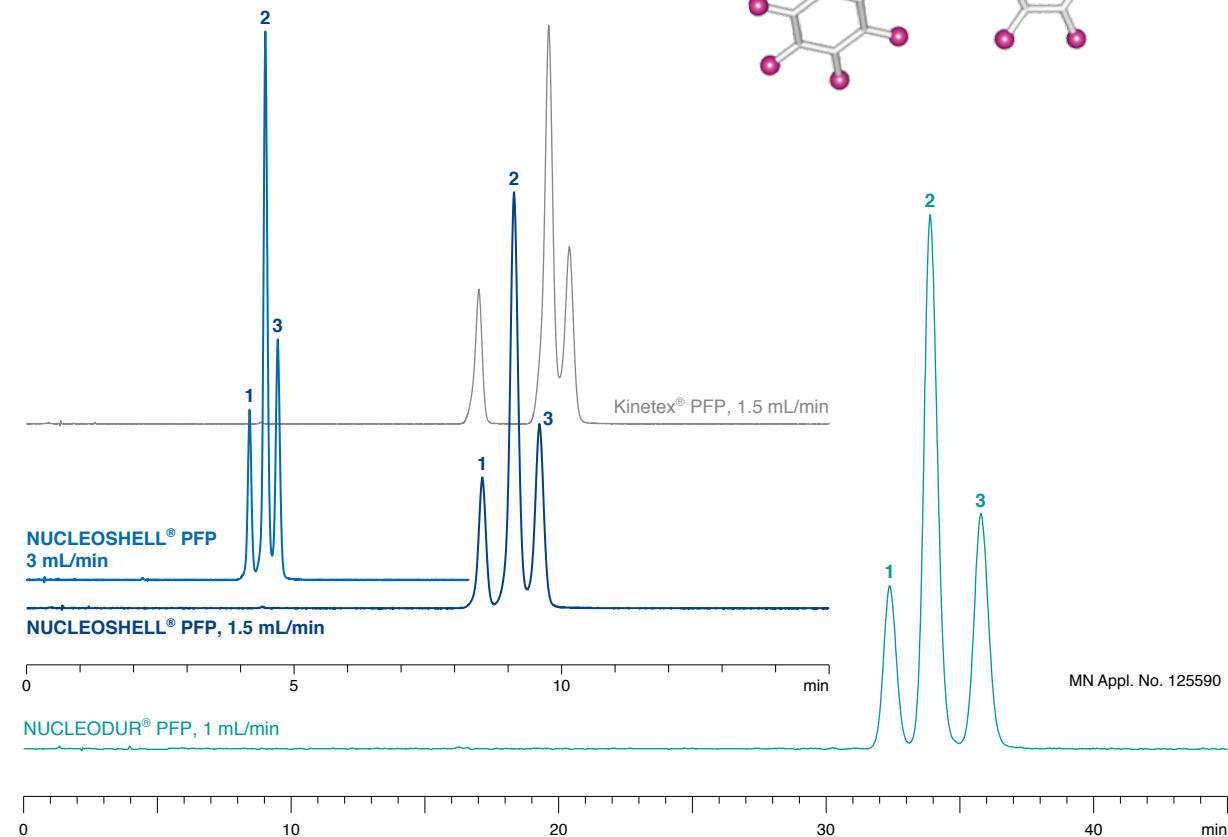


MN Appl. No. 125560

## Methylacetophenones

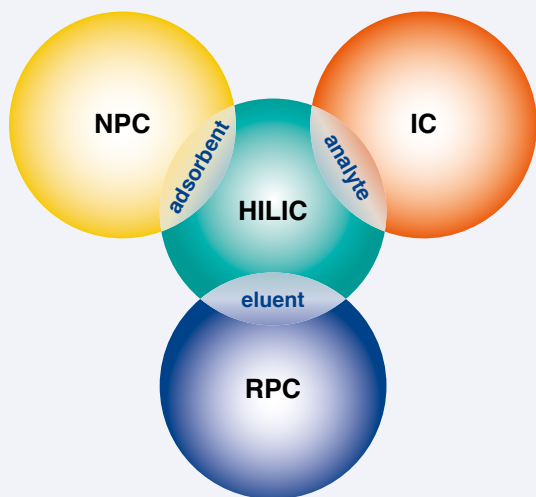
Columns: **100 x 4.6 mm NUCLEOSHELL® PFP, 2.7 µm**  
**250 x 4 mm NUCLEODUR® PFP, 5 µm**  
**100 x 4.6 mm Kinetex® 2.6 µm PFP**  
 Eluent: methanol – water (35:65, v/v)  
 Flow rate: **1.5 mL/min, 3 mL/min, 1 mL/min, 1.5 mL/min**  
 Temperature: 35 °C  
 Detection: UV, 254 nm

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





# NUCLEOSHELL® HILIC



## Key features:

- Based on core-shell particle technology for fast and efficient HPLC
- Ideal for reproducible and stable chromatography of highly polar analytes
- Very short column equilibration times

## Technical characteristics:

Ammonium - sulfonic acid modified silica;  
pore size 90 Å,  
particle size 2.7 µm;  
carbon content 1.3 %;  
pH stability 2–8.5; suitable for LC/MS

## Recommended application:

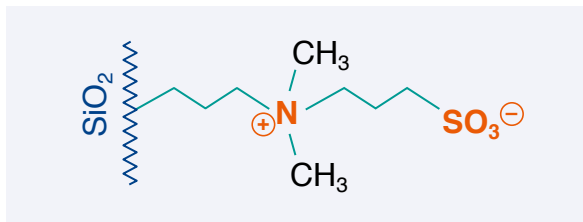
Hydrophilic compounds such as organic polar acids and bases, polar natural compounds, nucleosides, oligonucleotides, amino acids, peptides, water soluble vitamins

## NUCLEOSHELL® HILIC

Hydrophilic interaction chromatography (HILIC) is a separation technique using polar stationary phases and organic-aqueous mobile phases. A minimum water content of at least 2% is indispensable to provide a permanent water layer between the adsorbent surface and the organic fraction of the mobile phase. The sample molecules become separated in a partition chromatography, in which polar analytes are more strongly retained than neutral, less hydrophilic compounds. Consequently, increasing the aqueous part in the mobile phase will diminish retention of the polar sample constituents. In this way HILIC behaves inverse to classical RP chromatography. The particular retention profile of HILIC enables the chromatography of very polar and often small molecules, which won't show any retention on C<sub>8</sub> or C<sub>18</sub> reversed phases.

### Ultra-fast separations at moderate back pressure

NUCLEOSHELL® HILIC is a core-shell technology based stationary phase with a covalently bonded 3-*N,N*-dimethylaminopropane sulfonic acid ligand (pat. pend.). The betaine character of the strong ion-exchanger results in full charge balancing and facilitates fast equilibration times.



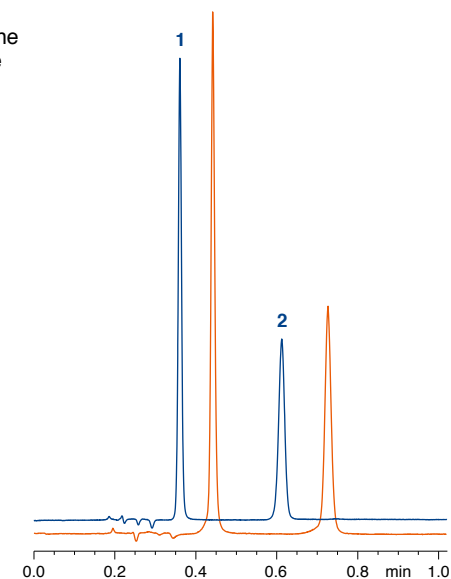
Good separation of polar compounds like the physiologically important substances creatine and creatinine can be achieved on NUCLEOSHELL® HILIC as well as on NUCLEODUR® HILIC, 1.8 µm at similar retention, but much lower back pressure.

### Separation of creatine and creatinine

Columns: **50 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
**50 x 4 mm NUCLEODUR® HILIC, 1.8 µm**  
Eluent: acetonitrile – 10 mmol/L ammonium acetate  
pH 4.0 (90:10, v/v)  
Flow rate: 1.7 mL/min  
Pressure: **129 bar**  
**180 bar**  
Temperature: 25 °C  
Detection: UV, 210 nm

#### Peaks:

1. Creatinine
2. Creatine

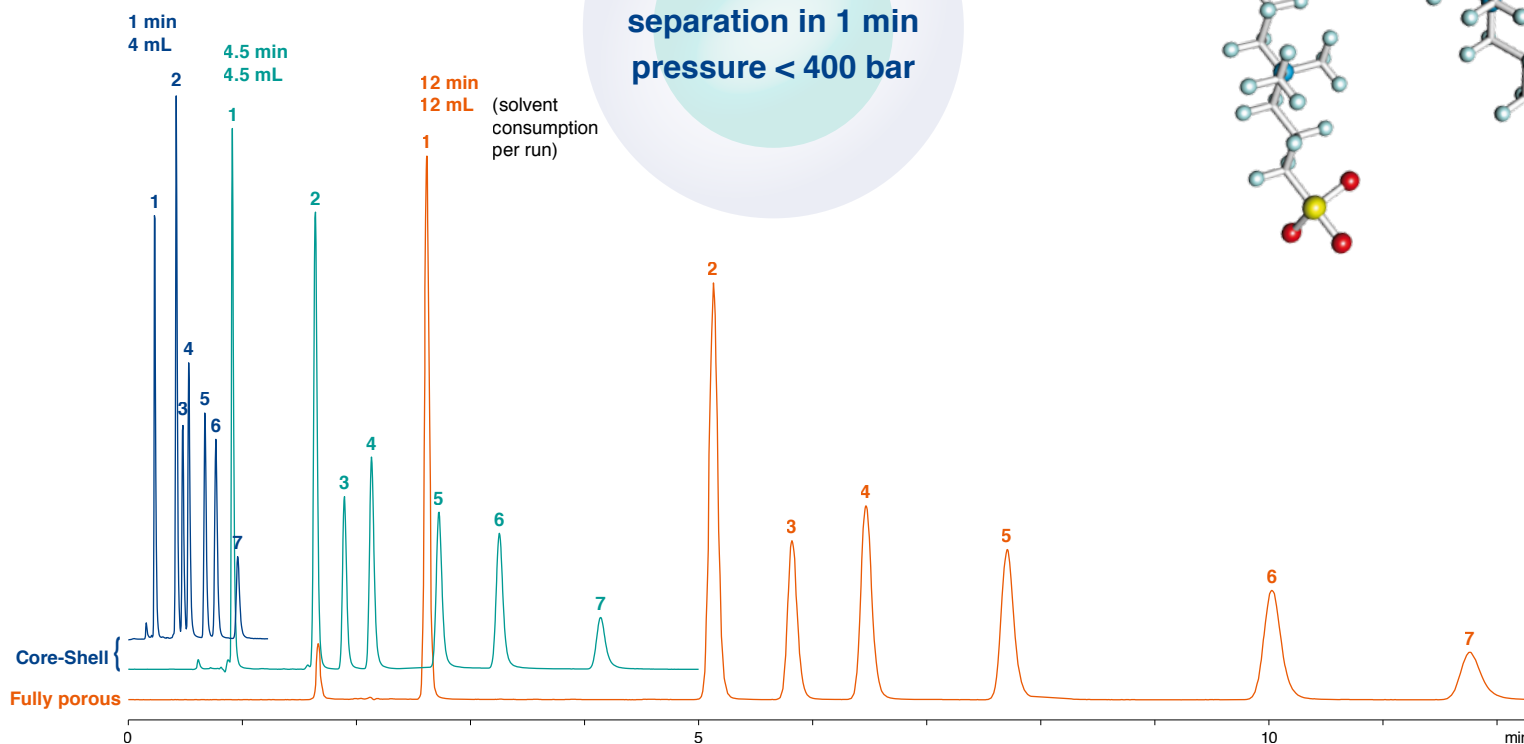


MN Appl. No. 124990

## Separation of catecholamines

Columns: **100 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
**100 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
**250 x 4 mm NUCLEODUR® HILIC, 3 µm**  
 Eluent: acetonitrile – 100 mmol/L ammonium formate pH 3.2 (80:20, v/v)  
 Flow rate: **4 mL/min, 1 mL/min, 1 mL/min**  
 Pressure: **395 bar, 95 bar, 116 bar**  
 Temperature: 25 °C  
 Detection: UV, 280 nm

- Peaks:**
1. DOPAC
  2. Serotonin
  3. Dopamine
  4. Epinephrine
  5. Norepinephrine
  6. DOPA
  7. DOPS



MN Appl. No. 125440

The chromatograms show the method transfer from a fully porous 3 µm HILIC phase to 2.7 µm core-shell silica with equal selectivity features. Run time has been cut

down to 1 min. Column back pressure remains modest < 400 bar, while solvent demand is reduced to less than 35%.

**NUCLEOSHELL® HILIC provides stable and reproducible chromatography, comprising all the benefits of a state-of-the-art core-shell silica.**

# Applications

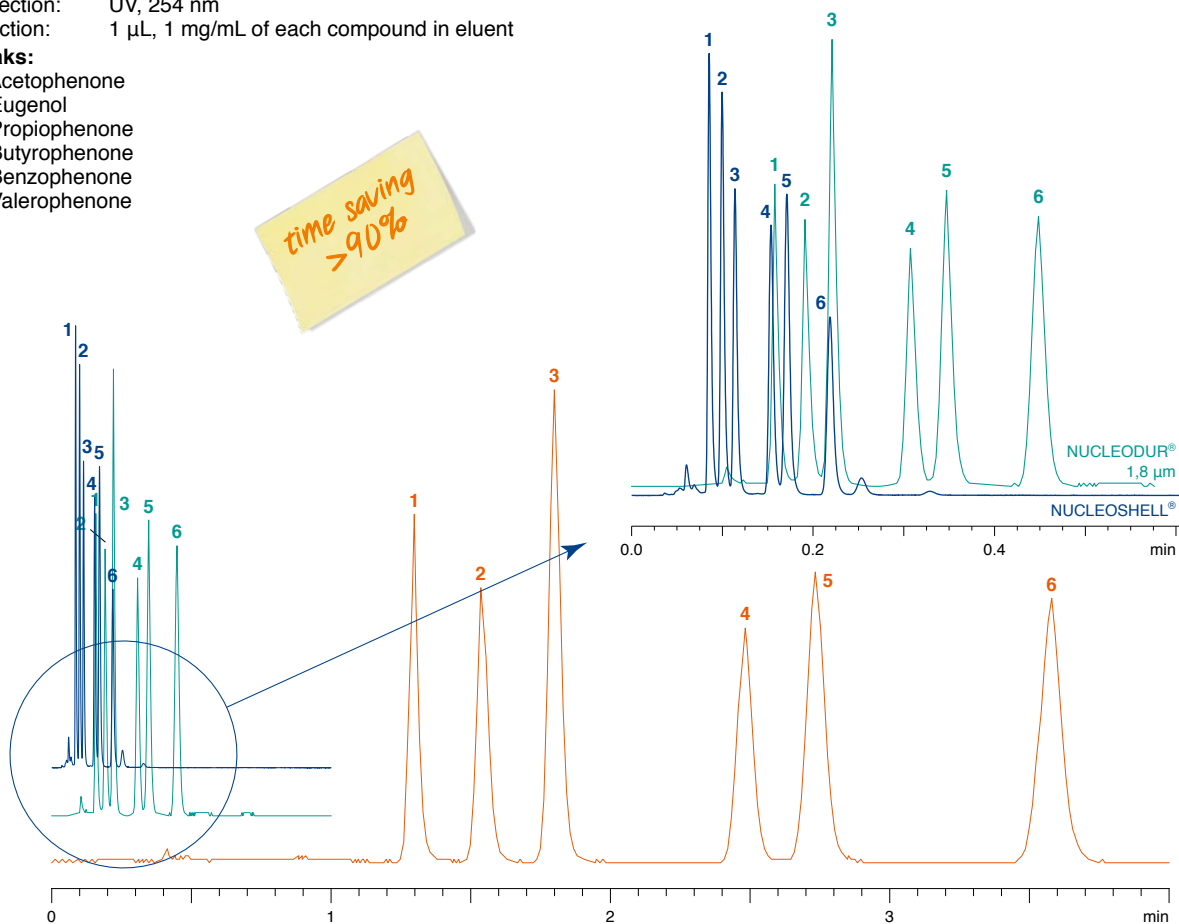
## Separation of ketones

Columns: 50 x 3 mm NUCLEOSHELL® RP 18, 2.7 µm  
 50 x 2 mm NUCLEODUR® C<sub>18</sub> Gravity, 1.8 µm  
 125 x 2 mm NUCLEODUR® C<sub>18</sub> Gravity, 5 µm  
 Eluent: acetonitrile – water (60:40, v/v)  
 Flow rate: 4 mL/min, 1.25 mL/min, 0.33 mL/min  
 Pressure: 540 bar, 774 bar, 89 bar  
 Temperature: 25 °C  
 Detection: UV, 254 nm  
 Injection: 1 µL, 1 mg/mL of each compound in eluent

### Peaks:

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

time saving  
>90%



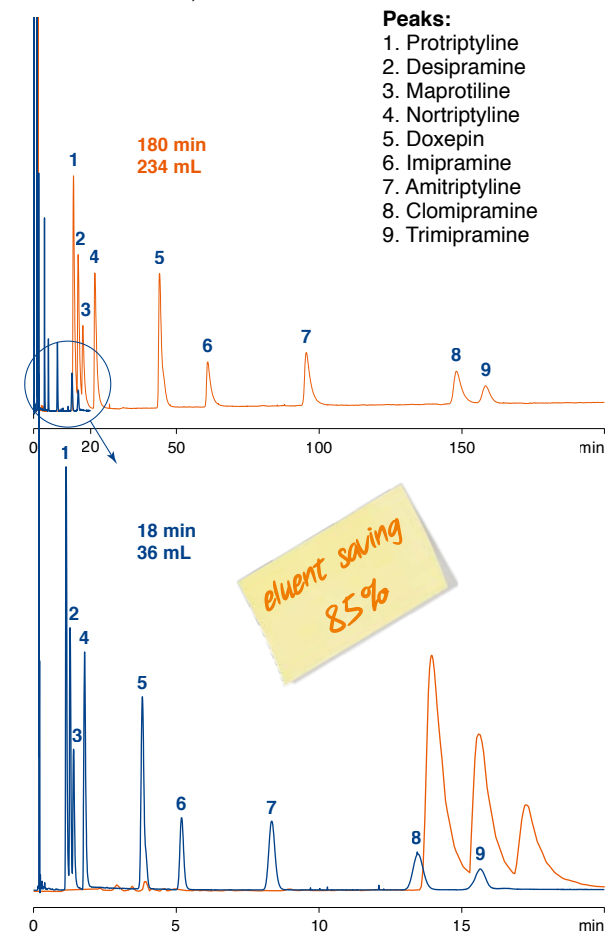
MN Appl. No. 124920

## Tricyclic antidepressants

Columns: 50 x 4.6 mm NUCLEOSHELL® RP 18, 2.7 µm  
 250 x 4.6 mm fully porous C<sub>18</sub>, 5 µm  
 Eluent: methanol – acetonitrile – 25 mmol/L KH<sub>2</sub>PO<sub>4</sub>  
 pH 7 (22.5:22.5:55, v/v)  
 Flow rate: 2 mL/min, 1.3 mL/min  
 Pressure: 224 bar, 190 bar  
 Temperature: 40 °C  
 Detection: UV, 220 nm

### Peaks:

1. Protriptyline
2. Desipramine
3. Maprotiline
4. Nortriptyline
5. Doxepin
6. Imipramine
7. Amitriptyline
8. Clomipramine
9. Trimipramine

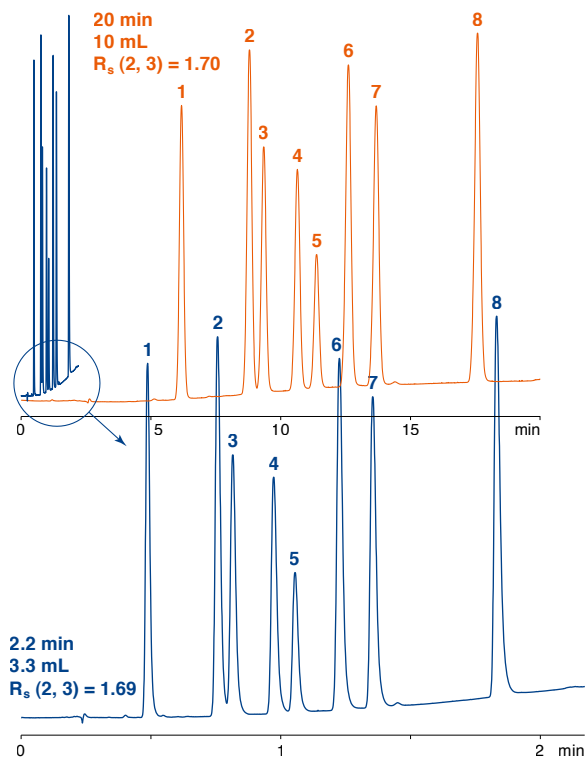


MN Appl. No. 125420

## Acidic pharmaceuticals

Columns: **50 x 4 mm NUCLEOSHELL® RP 18, 2.7 µm**  
 150 x 4 mm fully porous C<sub>18</sub>, 5 µm  
 Eluent: A) acetonitrile, B) 25 mmol/L KH<sub>2</sub>PO<sub>4</sub> pH 7,  
 25–40 % A in 2.2 min, 25–40 % A in 20 min  
 Flow rate: 1.5 mL/min, 0.5 mL/min  
 Pressure: 219 bar, 92 bar  
 Temperature: 20 °C  
 Detection: UV, 215 nm

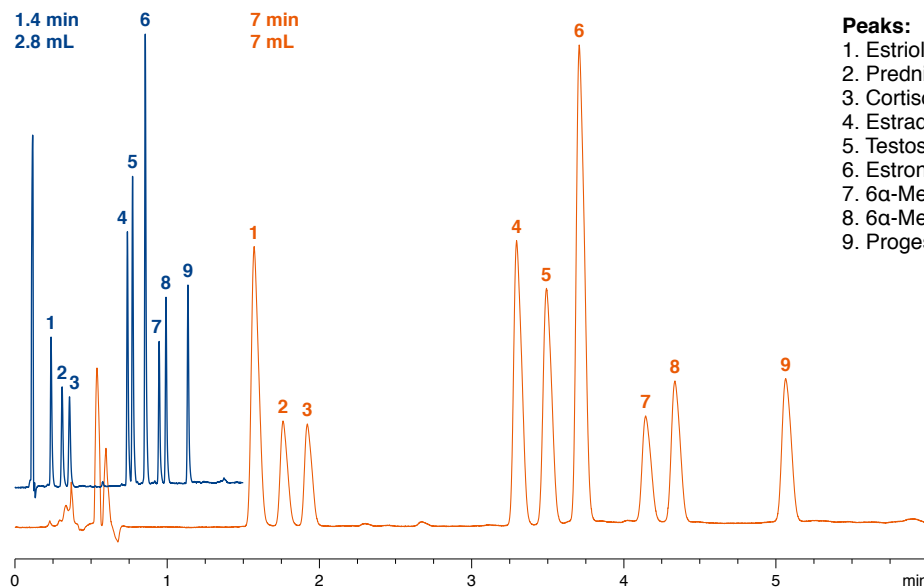
- Peaks:**
- |                 |                      |
|-----------------|----------------------|
| 1. Ketoprofen   | 5. Ibuprofen         |
| 2. Fenoprop     | 7. Carprofen         |
| 3. Fenoprofen   | 8. Diclofenac        |
| 4. Flurbiprofen | 9. Meclufenamic acid |



MN Appl. No. 125430

## Separation of steroids

Columns: **50 x 3 mm NUCLEOSHELL® RP 18, 2.7 µm**  
 125 x 3 mm NUCLEODUR® C<sub>18</sub> Gravity, 3 µm  
 Eluent: A) acetonitrile, B) water  
 30–80 % A in 1 min (0.4 min 80 % A)  
 30–80 % A in 5 min (2 min 80 % A)  
 Flow rate: 2 mL/min  
 1 mL/min  
 Pressure: 350 bar  
 280 bar  
 Temperature: 25 °C  
 Detection: UV, 240 nm  
 Injection: 1 µL, 1 mg/mL of each compound in eluent



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

MN Appl. No. 124930

*up to  
90% time saving  
66% solvent saving*

# Applications

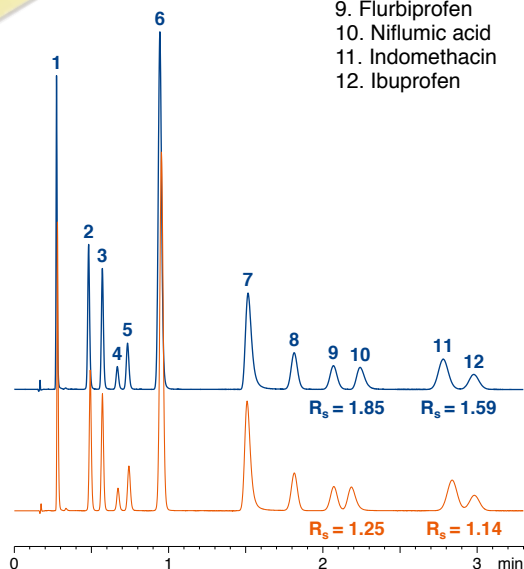
## Non-steroidal anti-inflammatory drugs

Columns: 50 x 4.6 mm each  
**NUCLEOSHELL® RP 18, 2.7 µm**  
**Ascentis® Express C<sub>18</sub>**  
 Eluent: acetonitrile – 20 mmol/L KH<sub>2</sub>PO<sub>4</sub> pH 2.5  
 (40:60, v/v)  
 Flow rate: 2.5 mL/min  
 Pressure: **268 bar, 281 bar**  
 Temperature: 22 °C  
 Detection: UV, 230 nm  
 Injection: 1 µL, 1 mg/mL of each compound in eluent

### 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

*good  
selectivity  
and resolution*



MN Appl. No. 124970

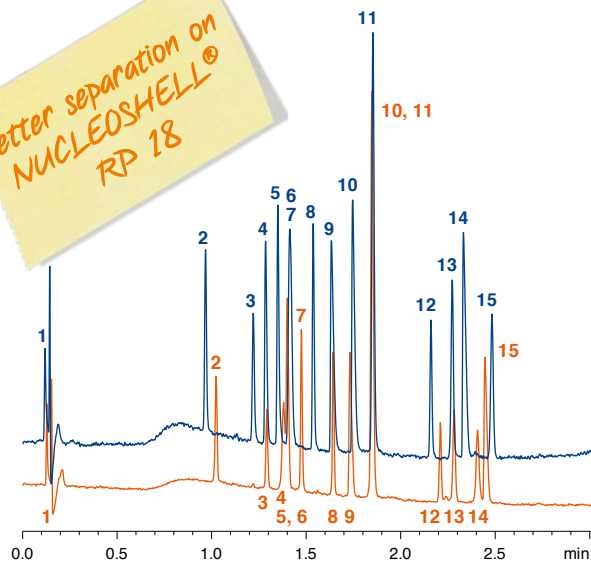
## Pharmaceuticals

Columns: 50 x 3 mm  
**NUCLEOSHELL® RP 18, 2.7 µm**  
**Kinetex® 2.6 µm C<sub>18</sub>**  
 Eluent: A) water + 0.1 % formic acid, B) acetonitrile +  
 0.1 % formic acid; 100 % A → 95 % A in 0.2 min  
 → 35 % A in 3.3 min  
 Flow rate: 2 mL/min  
 Temperature: 30 °C  
 Detection: UV, 245 nm  
 Injection: 1 µL

### Peaks:

- |                   |                                    |
|-------------------|------------------------------------|
| 1. Pyridine       | 9. Chlorpheniramine                |
| 2. Acetaminophen  | 10. Triprolidine                   |
| 3. Sulfathiazole  | 11. Salicylic acid                 |
| 4. Pindolol       | 12. Prednisolone                   |
| 5. Quinidine      | 13. 3-Methyl-4-nitrobenzoic acid   |
| 6. Benzyl alcohol | 14. Nortriptyline                  |
| 7. Phenol         | 15. 2-Hydroxy-5-methylbenzaldehyde |
| 8. Acebutolol     |                                    |

*better separation on  
NUCLEOSHELL®  
RP 18*



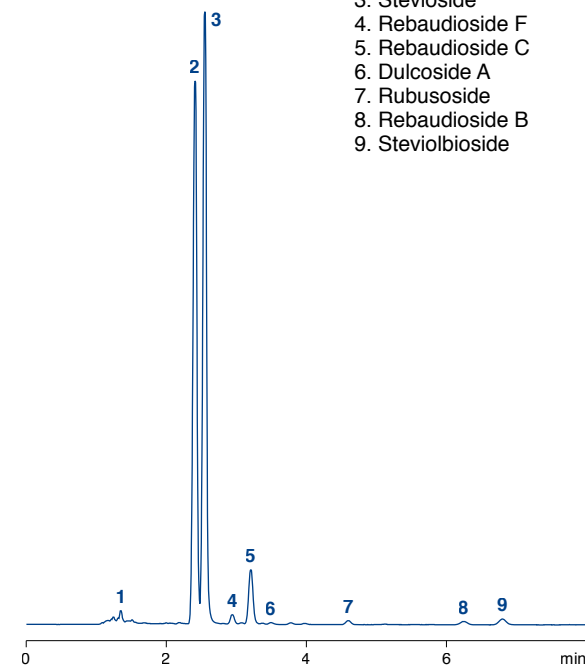
MN Appl. No. 126680

## Steviol glycosides

Column: **150 x 4.6 mm**  
**NUCLEOSHELL® RP 18, 2.7 µm**  
 Eluent: acetonitrile – 10 mmol/L NaH<sub>2</sub>PO<sub>4</sub> pH 2.6  
 (32:68, v/v)  
 Flow rate: 1.0 mL/min  
 Temperature: 40 °C  
 Detection: UV, 210 nm  
 Injection: 5 µL

### Peaks:

1. Rebaudioside D
2. Rebaudioside A
3. Stevioside
4. Rebaudioside F
5. Rebaudioside C
6. Dulcoside A
7. Rubusoside
8. Rebaudioside B
9. Steviolbioside



MN Appl. No. 125621



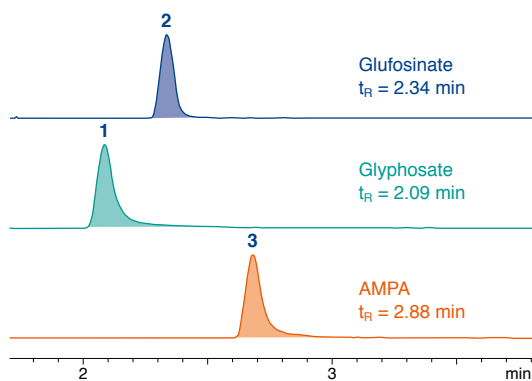
## Phosphonic acid herbicides

Columns: **100 x 2 mm**  
**NUCLEOSHELL® RP 18, 2.7 µm**

Eluent: A) acetonitrile,  
 B) 50 mmol/L ammonium acetate;  
 5–50% A in 3.7 min,  
 50–95% A in 0.6 min (2 min 95% A),  
 95–5% A in 0.5 min (2 min 5% A)

Flow rate: 0.5 mL/min  
 Temperature: 30 °C  
 Detection: MS  
 Injection: 5 µL

**Peaks:**  
 1. Glyphosate (167 ng/mL)  
 2. Glufosinate (16.7 ng/mL)  
 3. AMPA (167 ng/mL)



Courtesy of KUDZU SCIENCE, Illkirch, France

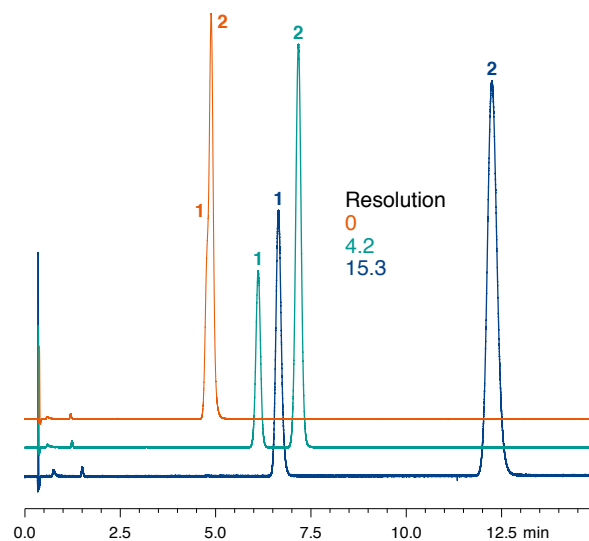
MN Appl. No. 126110

## Comparison of steric selectivity

Columns: 50 x 3 mm each  
**NUCLEOSHELL® RP 18plus, 2.7 µm**  
**NUCLEOSHELL® RP 18, 2.7 µm**  
**NUCLEODUR® C<sub>18</sub> Isis, 1.8 µm**

Eluent: methanol – water (70:30, v/v)  
 Flow rate: 0.56 mL/min  
 Temperature: 40 °C  
 Detection: UV, 254 nm  
 Injection: 2.0 µL

**Peaks:**  
 1. *o*-Terphenyl  
 2. Triphenylene



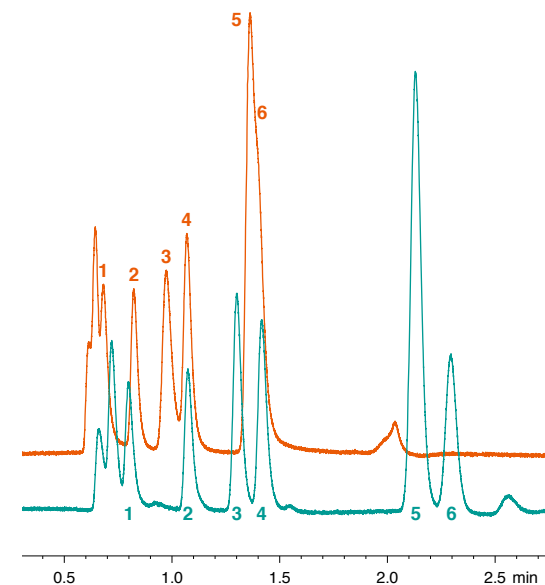
MN Appl. No. 126690

## Organic acids

Columns: 50 x 3 mm  
**NUCLEOSHELL® RP 18plus, 2.7 µm**  
**NUCLEOSHELL® RP 18, 2.7 µm**

Eluent: 20 mmol/L KH<sub>2</sub>PO<sub>4</sub> pH 2.6  
 Flow rate: 0.3 mL/min  
 Temperature: 20 °C  
 Detection: UV, 215 nm  
 Injection: 1 µL

**Peaks:**  
 1. Tartaric acid  
 2. Malic acid  
 3. Acetic acid  
 4. Lactic acid  
 5. Fumaric acid  
 6. Citric acid



MN Appl. No. 126700

# Applications

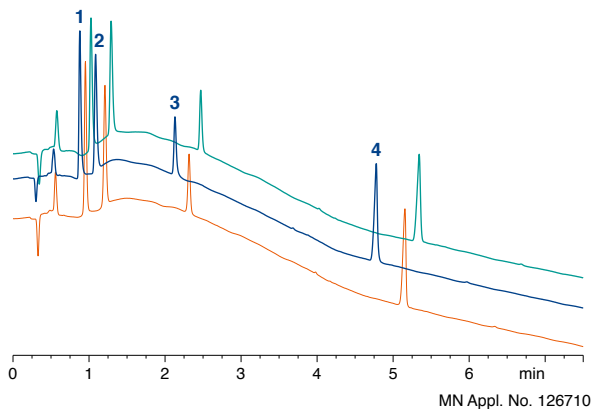
## Sweeteners

Columns: 50 x 3 mm  
**NUCLEOSHELL® RP 18plus, 2.7 µm**  
**NUCLEOSHELL® RP 18, 2.7 µm**  
**Kinetex® 2.6 µm C18**

Eluent: A) 21 mmol/L triethylammonium acetate  
 pH 4.5, B) acetonitrile; 10–30% B in 3 min,  
 30–39% B in 3.5 min

Flow rate: 0.45 mL/min  
 Temperature: 25 °C  
 Detection: UV, 220 nm  
 Injection: 1 µL

**Peaks:**  
 1. Acesulfame K  
 2. Saccharin  
 3. Aspartame  
 4. Stevioside



## β-Lactam antibiotics

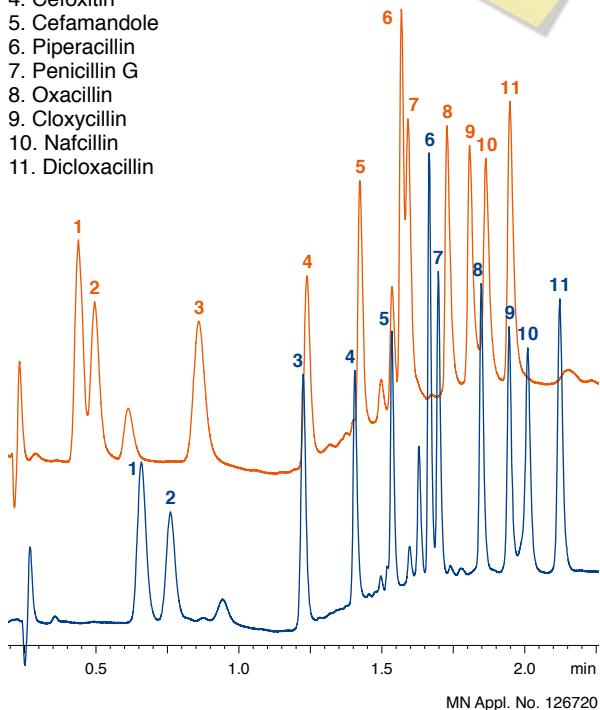
Columns: 50 x 2 mm  
**NUCLEOSHELL® RP 18plus, 2.7 µm**  
**NUCLEOSHELL® RP 18, 2.7 µm**  
**NUCLEOSHELL® RP 18, 2.7 µm**

Eluent: A) 20 mmol/L KH<sub>2</sub>PO<sub>4</sub> pH 3.5, B) acetonitrile;  
 90% A (0.25 min) → 50% A in 0.75 min (2 min)

Flow rate: 0.5 mL/min  
 Temperature: 20 °C  
 Detection: UV, 220 nm  
 Injection: 1 µL

**Peaks:**  
 1. Ampicillin  
 2. Cephalaxine  
 3. Cefotaxime  
 4. Cefoxitin  
 5. Cefamandole  
 6. Piperacillin  
 7. Penicillin G  
 8. Oxacillin  
 9. Cloxacillin  
 10. Nafcillin  
 11. Dicloxacillin

*better separation  
 due to higher  
 retention*



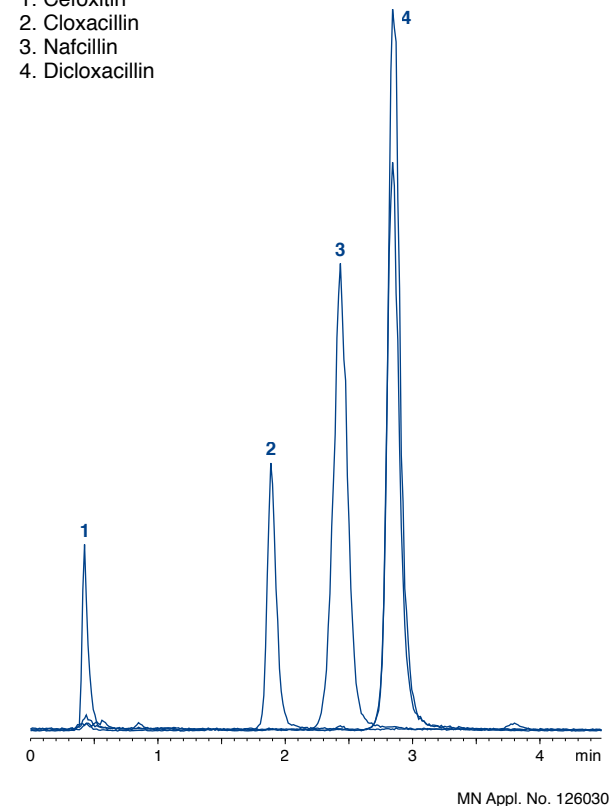
## β-Lactam antibiotics

Column: 50 x 2 mm  
**NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm**

Eluent: methanol – 10 mmol/L ammonium formate,  
 pH 3 (50:50, v/v)

Flow rate: 0.45 mL/min  
 Temperature: 40 °C  
 Detection: MS  
 Injection: 1 µL

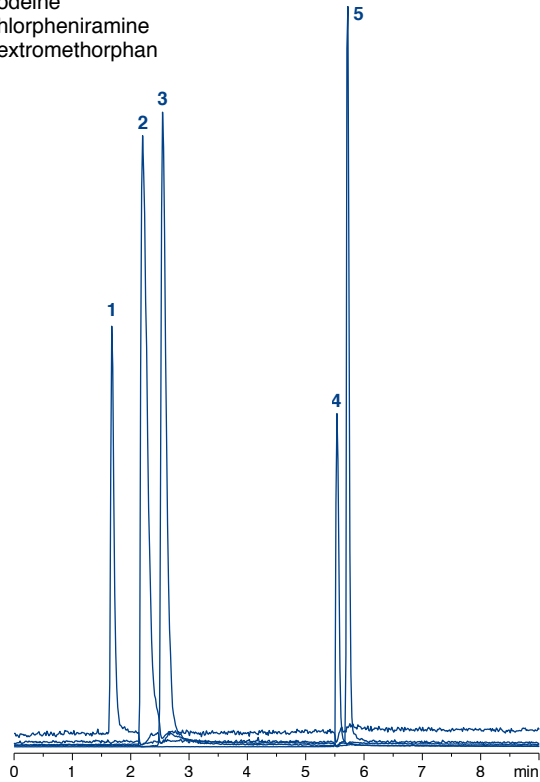
**Peaks:**  
 1. Cefoxitin  
 2. Cloxacillin  
 3. Nafcillin  
 4. Dicloxacillin



## Antihistamines

Column: **100 x 3 mm**  
**NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm**  
 Eluent: A) methanol,  
 B) 10 mmol/L ammonium formate, pH 2.8;  
 17.5% A (2.5 min) → 65% A in 1.5 min →  
 75% A in 1.5 min (4.5 min 75% A)  
 Flow rate: 0.6 mL/min  
 Temperature: 40 °C  
 Detection: MS  
 Injection: 0.5 µL

**Peaks:**  
 1. 4-Acetaminophenol  
 2. Pseudoephedrine  
 3. Codeine  
 4. Chlorpheniramine  
 5. Dextromethorphan

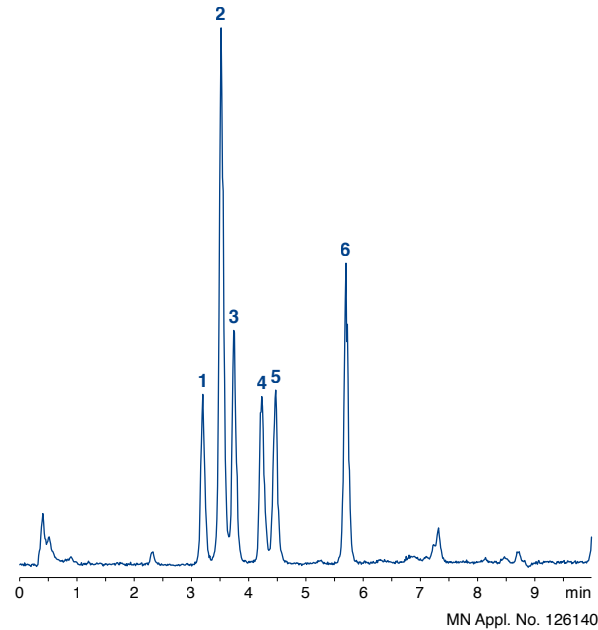


MN Appl. No. 125950

## Benzodiazepines

Column: **50 x 2 mm**  
**NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm**  
 Eluent: A) acetonitrile,  
 B) 20 mmol/L ammonium formate, pH 6.4;  
 25–55% A in 10 min  
 Flow rate: 0.33 mL/min  
 Temperature: 25 °C  
 Detection: MS  
 Injection: 2.5 µL

**Peaks:**  
 1. Oxazepam  
 2. Chlordiazepoxide  
 3. Alprazolam  
 4. Trazodone  
 5. Nordiazepam  
 6. Diazepam

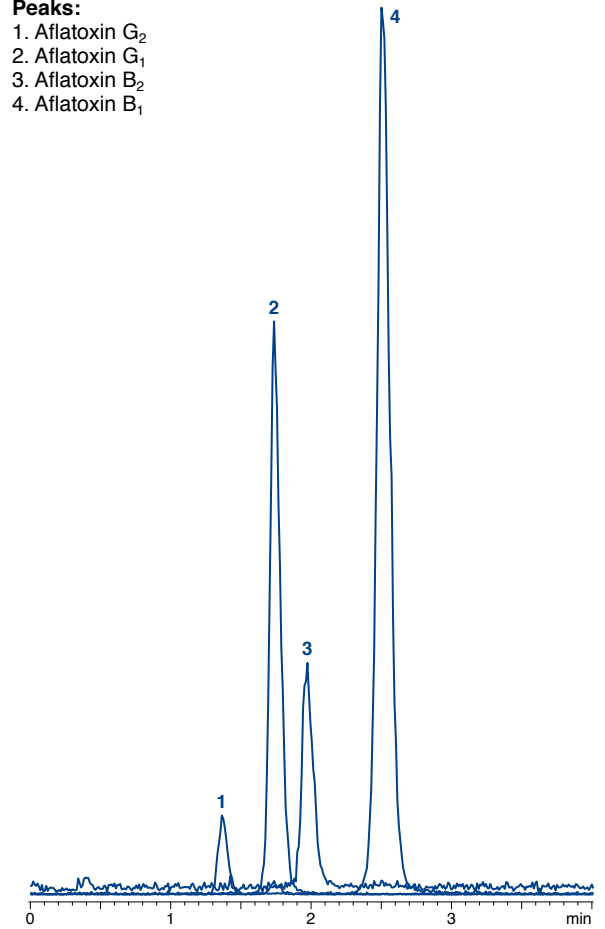


MN Appl. No. 126140

## Aflatoxins

Column: **50 x 2 mm NUCLEOSHELL® PFP, 2.7 µm**  
 Eluent: methanol – 10 mmol/L ammonium acetate  
 (45:55, v/v)  
 Flow rate: 0.33 mL/min  
 Temperature: 25 °C  
 Detection: MS  
 Injection: 0.1 ng each

**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>



MN Appl. No. 125600

# Applications

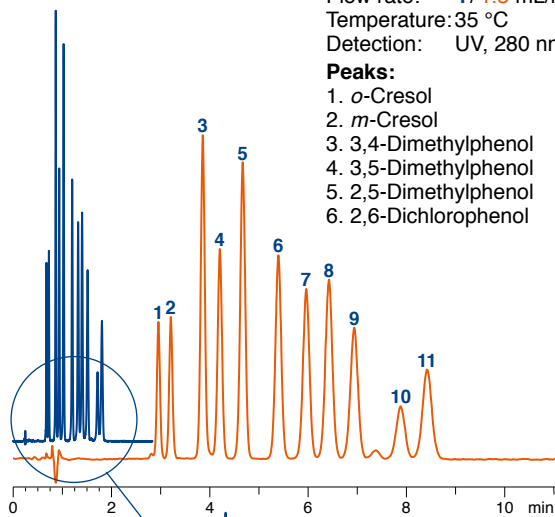
## Phenols

Columns: **100 x 4.6 mm NUCLEOSHELL® PFP, 2.7 µm**  
**100 x 4.6 mm NUCLEODUR® PFP, 5 µm**  
 Eluent: acetonitrile + 0.1 % formic acid – 0.1 % formic acid (35:65, v/v)

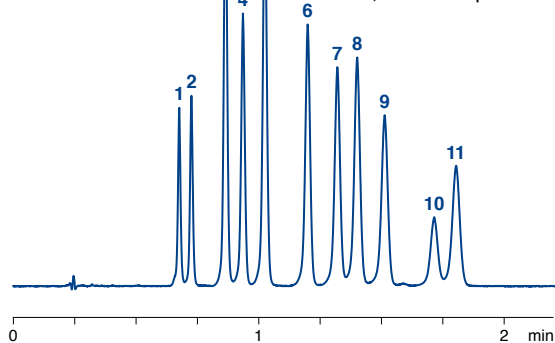
Flow rate: **4 / 1.3 mL/min**  
 Temperature: 35 °C  
 Detection: UV, 280 nm

### 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-Dichlorophenol



MN Appl. No. 125570

## Beta- and dexamethasone

Columns: **50 x 4 mm NUCLEOSHELL® PFP, 2.7 µm**  
**100 x 4.6 mm NUCLEODUR® PFP, 5 µm**

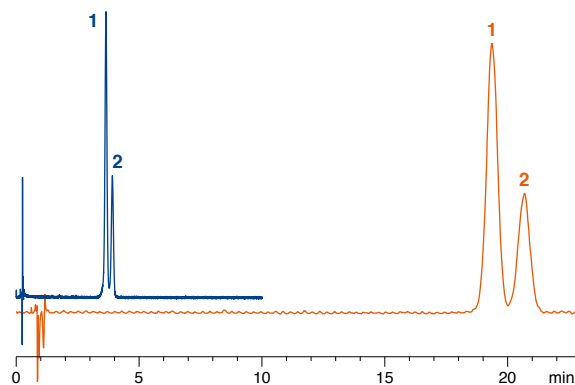
Eluent: acetonitrile – water (20:80, v/v)

Flow rate: **1.5 mL/min**  
**1.3 mL/min**

Temperature: 30 °C  
 Detection: UV, 260 nm

### Peaks:

1. Betamethasone
2. Dexamethasone



MN Appl. No. 125580

## Water-soluble vitamins

Columns: 100 x 3 mm  
**NUCLEOSHELL® RP 18plus, 2.7 µm**  
**NUCLEOSHELL® RP 18, 2.7 µm**

Eluent: A) 20 mmol/L KH<sub>2</sub>PO<sub>4</sub> pH 3, B) acetonitrile – methanol (30:70, v/v); 100% A (2 min) → 70% A in 1 min (12 min)

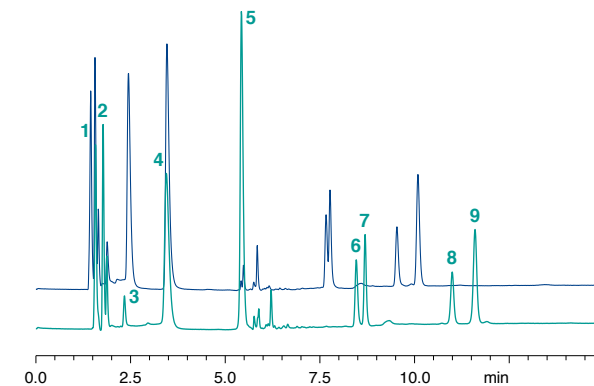
Flow rate: 0.25 mL/min  
 Temperature: 15 °C

Detection: UV, 220 nm  
 Injection: 1 µL

### Peaks:

1. Pyridoxamine
2. Thiamine (vitamin B<sub>1</sub>)
3. Ascorbic acid (vitamin C)
4. Pyridoxal
5. Pyridoxine (vitamin B<sub>6</sub>)
6. Folic acid
7. Cyanocobalamine (vitamin B<sub>12</sub>)
8. Riboflavin
9. Biotin

*Higher retention on RP 18plus due to polar selectivity*



MN Appl. No. 126730

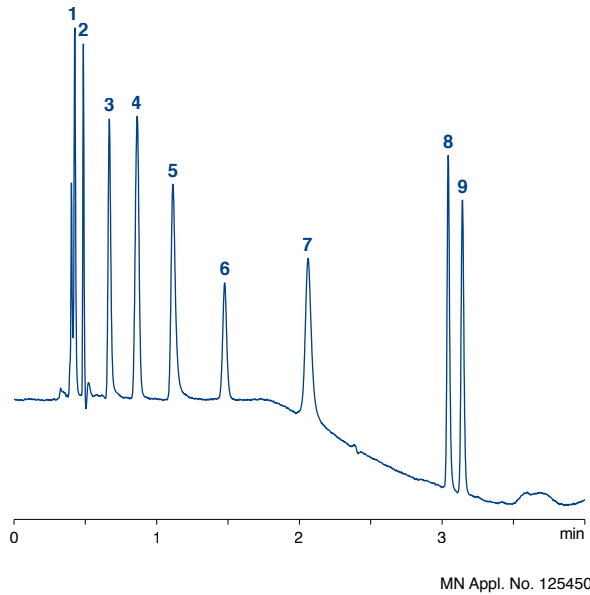
## Water-soluble vitamins

Column: **100 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
 Eluent: A) acetonitrile – 100 mmol/L ammonium acetate pH 3.2 (90:10, v/v), B) water; 4% B (1 min) → 20% B in 1.6 min (0.7 min 20% B)

Flow rate: 2 mL/min  
 Pressure: 218 bar  
 Temperature: 25 °C  
 Detection: UV, 260 nm

### Peaks:

1. PABA (*p*-aminobenzoic acid)
2. Nicotinamide
3. Vitamin B<sub>6</sub> (pyridoxine)
4. Riboflavin
5. Nicotinic acid
6. Vitamin C (ascorbic acid)
7. Vitamin B<sub>1</sub> (thiamine)
8. Folic acid
9. Vitamin B<sub>12</sub> (cyanocobalamin)



## Metformin

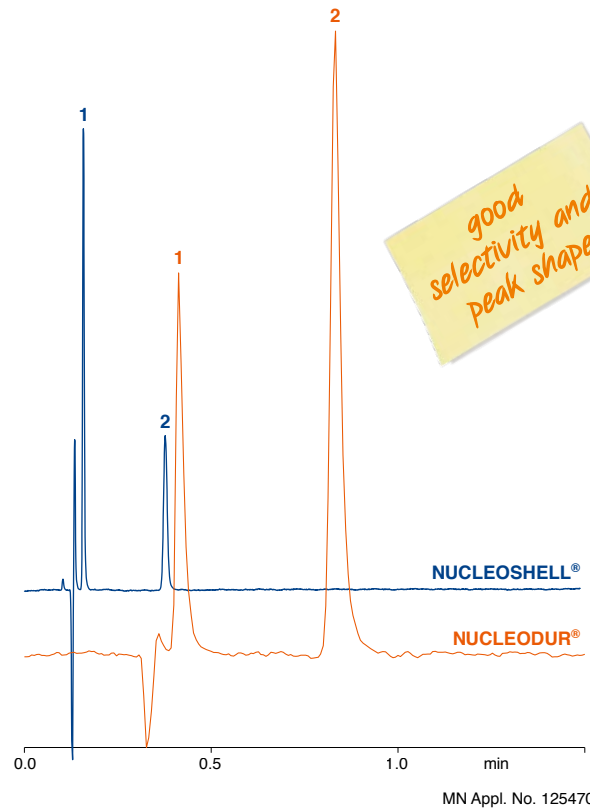
Columns: **50 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
**50 x 4 mm NUCLEODUR® HILIC, 1.8 µm**  
 Eluent: acetonitrile – 10 mmol/L ammonium acetate pH 3.2 (75:25, v/v)

Flow rate: **3 mL/min**  
**1.5 mL/min**  
 Pressure: **202 bar**  
**167 bar**

Temperature: 25 °C  
 Detection: UV, 218 nm

### Peaks:

1. Dicyandiamide
2. Metformin



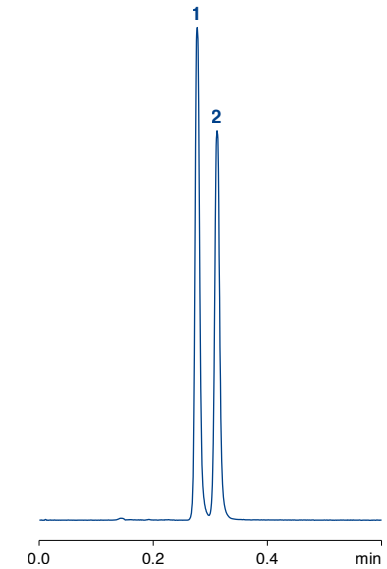
## 5-Fluorouracil

Column: **50 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
 Eluent: acetonitrile – 10 mmol/L ammonium acetate (95:5, v/v)

Flow rate: 2.5 mL/min  
 Pressure: 119 bar  
 Temperature: 25 °C  
 Detection: UV, 254 nm

### Peaks:

1. 5-Fluorouracil
2. Uracil



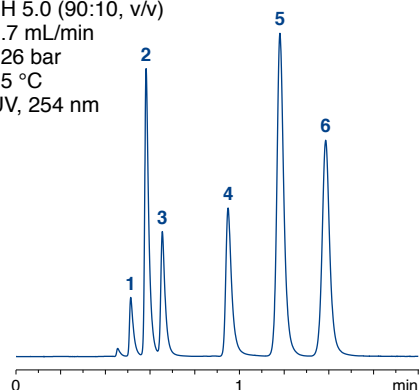


# Applications · Guard column system

## Analysis of an energy drink

Column: **100 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
Eluent: acetonitrile – 100 mmol/L ammonium acetate pH 5.0 (90:10, v/v)  
Flow rate: 1.7 mL/min  
Pressure: 126 bar  
Temperature: 35 °C  
Detection: UV, 254 nm

**Peaks:**  
1. Caffeine  
2. Niacinamide  
3. Pyridoxine  
4. Benzoic acid  
5. Sorbic acid  
6. Riboflavin

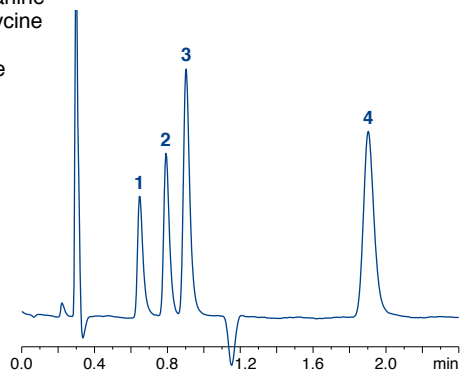


MN Appl. No. 125010

## Amino acids

Column: **50 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
Eluent: acetonitrile – 100 mmol/L ammonium acetate pH 4.0 (80:20, v/v)  
Flow rate: 1.5 mL/min  
Pressure: 105 bar  
Temperature: 25 °C  
Detection: UV, 215 nm

**Peaks:**  
1. Phenylalanine  
2. Phenylglycine  
3. Tyrosine  
4. Histamine

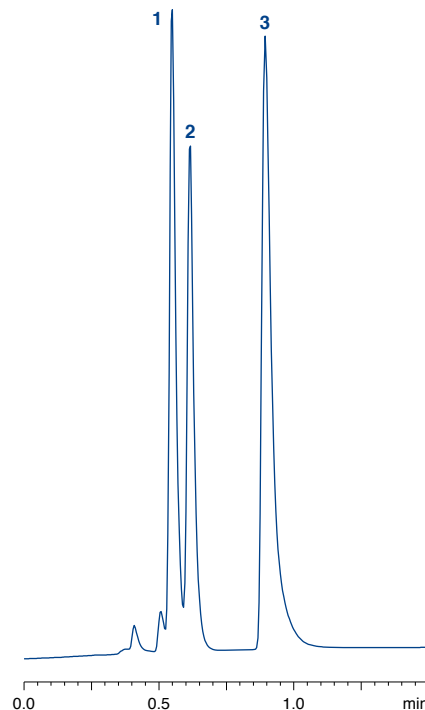


MN Appl. No. 125000

## Acrylamide and analogs

Column: **50 x 4 mm NUCLEOSHELL® HILIC, 2.7 µm**  
Eluent: acetonitrile – 0.1 % formic acid in water (98:2, v/v)  
Flow rate: 1.0 mL/min  
Temperature: 25 °C  
Detection: UV, 200 nm  
Injection: 1 µL, 1 mg/mL of each compound in eluent

**Peaks:**  
1. Acrylamide  
2. Methacrylamide  
3. Methacrylic acid



MN Appl. No. 125160

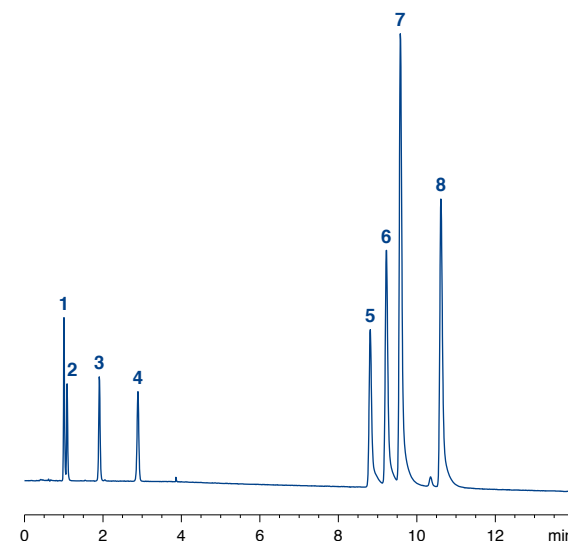
## Nucleotides

Column: **100 x 4.6 mm NUCLEOSHELL® HILIC, 2.7 µm**

Eluents: A) acetonitrile;  
B) 100 mmol/L ammonium acetate pH 5.35;  
87.5–60 % A in 18 min

Flow rate: 2.2 mL/min  
Temperature: 30 °C  
Detection: UV, 259 nm  
Injection: 1 µL, 1 mg/mL of each compound in eluent

**Peaks:**  
1. Uridine  
2. Adenosine  
3. Cytidine  
4. Cyclic adenosine monophosphate  
5. Uridine monophosphate  
6. Adenosine monophosphate  
7. Inosine monophosphate  
8. Cytidine monophosphate



MN Appl. No. 125200

## Column Protection System

Innovative and universal screw-on guard column holder system

Suitable for all analytical HPLC columns with 1/16" fittings



- Cartridges filled with specified NUCLEOSHELL®, NUCLEODUR®, and NUCLEOSIL® 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 available for all HPLC columns under RP conditions

## Content of the Column Protection System



Description	REF
<b>Column Protection System</b>	<b>718966</b>
<b>Details</b>	<b>Content</b>
Cartridge Holder	1
Replacement capillaries (0.12 mm ID)	2
Ferrules	3
Wrenches	2
Manual	1

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

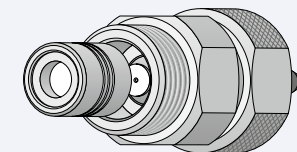
Description	Pack of	REF
Ferrules	5	718967
Replacement connector including O-ring	1	718968
Stainless steel capillaries 0.12 mm ID, nuts and metal ferrules	3	718969
Stainless steel capillaries 0.18 mm ID (for higher flow rates), nuts and metal ferrules	3	718971
Wrench (size 12 and 14 mm)	1	718970
EC 4/2 UNIVERSAL RP guard column (for main columns with 2 mm ID)	3	728777.20
EC 4/2 UNIVERSAL RP guard column (for main columns with 2 mm ID), value pack	9	728778.20
EC 4/3 UNIVERSAL RP guard column (for main columns with 3, 4 and 4.6 mm ID)	3	728777.30
EC 4/3 UNIVERSAL RP guard column (for main columns with 3, 4 and 4.6 mm ID), value pack	9	728778.30

## Contamination check

**The cartridge is fitted with a special filter membrane.**

If the silver membrane is contaminated (bright or dark discoloration), it is advisable to replace the cartridge.

If the contaminants are colorless, replace the cartridge as soon as the pressure rises or the chromatographic performance decreases.



# Packed columns · Technical information

## EC analytical columns

All phases: pore size 90 Å, eluent in column CH<sub>3</sub>CN – H<sub>2</sub>O

Length →	50 mm	100 mm	150 mm	250 mm	EC guard columns*
<b>NUCLEOSHELL® RP 18, 2.7 µm</b> octadecyl modification, particle size 2.7 µm, multi-encapped, 7.8 % C					
2 mm ID	763132.20	763134.20	763136.20		4 x 2 mm: 763138.20
3 mm ID	763132.30	763134.30	763136.30		4 x 3 mm: 763138.30
4 mm ID	763132.40	763134.40	763136.40		4 x 3 mm: 763138.30
4.6 mm ID	763132.46	763134.46	763136.46		4 x 3 mm: 763138.30

<b>NUCLEOSHELL® RP 18, 5 µm</b> octadecyl modification, particle size 5 µm, multi-encapped, 6.1 % C					
2 mm ID	763152.20	763154.20	763156.20	763157.20	4 x 2 mm: 763158.20
3 mm ID	763152.30	763154.30	763156.30	763157.30	4 x 3 mm: 763158.30
4 mm ID	763152.40	763154.40	763156.40	763157.40	4 x 3 mm: 763158.30
4.6 mm ID	763152.46	763154.46	763156.46	763157.46	4 x 3 mm: 763158.30

<b>NUCLEOSHELL® RP 18plus, 2.7 µm</b> polar octadecyl modification, particle size 2.7 µm, multi-encapped, 5.7 % C					
2 mm ID	763232.20	763234.20	763236.20		4 x 2 mm: 763238.20
3 mm ID	763232.30	763234.30	763236.30		4 x 3 mm: 763238.30
4 mm ID	763232.40	763234.40	763236.40		4 x 3 mm: 763238.30
4.6 mm ID	763232.46	763234.46	763236.46		4 x 3 mm: 763238.30

<b>NUCLEOSHELL® RP 18plus, 5 µm</b> polar octadecyl modification, particle size 5 µm, multi-encapped, 4.4 % C					
2 mm ID	763252.20	763254.20	763256.20	763257.20	4 x 2 mm: 763258.20
3 mm ID	763252.30	763254.30	763256.30	763257.30	4 x 3 mm: 763258.30
4 mm ID	763252.40	763254.40	763256.40	763257.40	4 x 3 mm: 763258.30
4.6 mm ID	763252.46	763254.46	763256.46	763257.46	4 x 3 mm: 763258.30

\* EC guard columns require the Column Protection System Cartridge Holder REF 718966 (see page 25).

EC columns in packs of 1, guard columns in packs of 3

## EC analytical columns

All phases: pore size 90 Å, particle size 2.7 µm; eluent in column CH<sub>3</sub>CN – H<sub>2</sub>O

Length →	50 mm	100 mm	150 mm	EC guard columns*
<b>NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm</b> phenyl-hexyl modification, multi-encapped, 4.5 % C				
2 mm ID	763732.20	763734.20	763736.20	4 x 2 mm: 763738.20
3 mm ID	763732.30	763734.30	763736.30	4 x 3 mm: 763738.30
4 mm ID	763732.40	763734.40	763736.40	4 x 3 mm: 763738.30
4.6 mm ID	763732.46	763734.46	763736.46	4 x 3 mm: 763738.30

<b>NUCLEOSHELL® PFP, 2.7 µm</b> pentafluorophenyl modification, multi-encapped, ~3 % C				
2 mm ID	763532.20	763534.20	763536.20	4 x 2 mm: 763538.20
3 mm ID	763532.30	763534.30	763536.30	4 x 3 mm: 763538.30
4 mm ID	763532.40	763534.40	763536.40	4 x 3 mm: 763538.30
4.6 mm ID	763532.46	763534.46	763536.46	4 x 3 mm: 763538.30

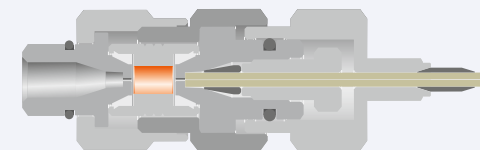
<b>NUCLEOSHELL® HILIC, 2.7 µm</b> ammonium – sulfonic acid modification, 1.3 % C				
2 mm ID	763332.20	763334.20	763336.20	4 x 2 mm: 763338.20
3 mm ID	763332.30	763334.30	763336.30	4 x 3 mm: 763338.30
4 mm ID	763332.40	763334.40	763336.40	4 x 3 mm: 763338.30
4.6 mm ID	763332.46	763334.46	763336.46	4 x 3 mm: 763338.30

\* EC guard columns require the Column Protection System Cartridge Holder REF 718966 (see page 25).  
EC columns in packs of 1, guard columns in packs of 3

## EC standard columns for analytical HPLC



- Analytical column system made of 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)  
EC column hardware guarantees pressure stability of 1200 bar - hence EC columns are suitable for U-HPLC applications (ultra fast HPLC) and all modern HPLC systems.
- As screw-on guard column system we recommend the **Column Protection System** used with EC guard column cartridges with 4 mm length (see page 25).

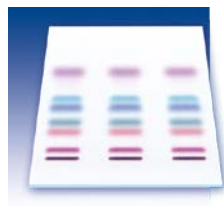




HPLC



GC



TLC



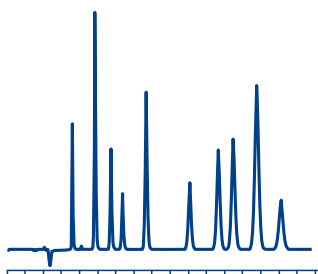
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