USP listing

USP specification of MN HPLC phases

Code	Specification	MN HPLC Phases	Page
USP L1	octadecyl silane chemically bonded to porous silica particles 1.5 to 10 µm diameter, or monolithic silica gel	NUCLEODUR® C_{18} ec	181
		NUCLEODUR® C_{18} Gravity	158
		NUCLEODUR® C_{18} Gravity-SB	162
		NUCLEODUR® C_{18} HTec	178
		NUCLEODUR® C_{18} Isis	164
		NUCLEODUR® C_{18} Pyramid	166
		NUCLEODUR® PolarTec	168
		NUCLEODUR® Sphinx RP	176
		NUCLEOSHELL® RP 18	200
		NUCLEOSHELL® RP 18plus	202
		NUCLEOSIL® C_{18}	220
		NUCLEOSIL® C_{18} AB	220
		NUCLEOSIL® C_{18} HD	220
		NUCLEOSIL® Nautilus	221
		NUCLEOSIL® C_{18} MPN	250
		NUCLEOSIL® C_{18} PPN	251
USP L3	porous silica particles, 1.5 to 10 µm diameter, or monolithic silica gel	NUCLEODUR® SiOH	190
		NUCLEOSIL® SiOH	230
USP L7	octyl silane chemically bonded to totally porous silica particles, 1.8 to 10 µm diameter	NUCLEODUR® C_8 ec	181
		NUCLEODUR® C_8 Gravity	158
		NUCLEOSIL® C_8	224
		NUCLEOSIL® C_8 HD	224
USP L8	an essentially monomolecular layer of aminopropyl silane chemically bonded to totally porous silica gel support, 1.5 to 10 µm diameter	NUCLEODUR® NH_2 / NH_2-RP	188
		NUCLEOSIL® Carbohydrate	254
		NUCLEOSIL® NH_2 / NH_2-RP	227
USP L9	irregular or spherical, totally porous silica gel having a chemically bonded, strongly acidic cation-exchange coating, 3 to 10 µm diameter	NUCLEOSIL® SA	229
USP L10	nitrile groups chemically bonded to porous silica particles, 1.5 to 10 µm diameter	NUCLEODUR® CN / CN-RP	186
		NUCLEOSIL® CN / CN-RP	228
USP L11	phenyl groups chemically bonded to porous silica particles, 1.5 to 10 µm diameter	NUCLEODUR® Phenyl-Hexyl	170
		NUCLEODUR® π^2	172
		NUCLEOSHELL® Phenyl-Hexyl	207
		NUCLEODUR® Sphinx RP	176
		NUCLEOSIL® C_6H_5	226
USP L14	silica gel having a chemically bonded, strongly basic quaternary ammonium anion-exchange coating, 5 to 10 µm diameter	NUCLEOSIL® SB	229
USP L16	dimethylsilane chemically bonded to porous silica particles, 5 to 10 µm diameter	NUCLEOSIL® C_2	225
USP L17	strong cation-exchange resin consisting of sulfonated cross-linked PS/DVB copolymer in the H form, 6 to 12 µm diameter	NUCLEOGEL® ION 300 OA	256
		NUCLEOGEL® SUGAR 810 H	255
USP L19	strong cation-exchange resin consisting of sulfonated cross-linked PS/DVB copolymer in the Ca form, 5 to 15 µm particle size	NUCLEOGEL® SUGAR 810 Ca	255
		NUCLEOGEL® SUGAR Ca	256
USP L20	dihydroxypropane groups chemically bonded to porous silica particles, 5 to 10 µm diameter	NUCLEOSIL® OH (Diol)	226
USP L21	a rigid, spherical styrene-divinylbenzene copolymer, 5 to 10 µm diameter	NUCLEOGEL® RP	252
USP L22	a cation-exchange resin made of porous polystyrene gel with sulfonic acid groups, about 10 µm in size	NUCLEOGEL® SCX	247
USP L23	an anion-exchange resin made of porous polymethacrylate or polyacrylate gel with quaternary ammonium groups, about 10 µm in size	NUCLEOGEL® SAX	247
USP L26	butyl silane chemically bonded to totally porous silica particles, 5 to 10 µm diameter	NUCLEODUR® C_4 ec	248
		NUCLEOSIL® C_4	225
		NUCLEOSIL® C_4 MPN	250
USP L32	a chiral ligand-exchange resin packing · L-proline copper complex covalently bonded to irregular shaped silica particles, 5 to 10 µm diameter	NUCLEOSIL® CHIRAL-1	242
USP L34	strong cation-exchange resin consisting of sulfonated cross-linked PS-DVB copolymer in the Pb form, 5 to 7 µm particle size	NUCLEOGEL® SUGAR Pb	256
USP L36	a 3,5-dinitrobenzoyl derivative of L-phenylglycine covalently bonded to 5 µm aminopropyl silica	NUCLEOSIL® CHIRAL-3	243
USP L40	cellulose tris-(3,5-dimethylphenylcarbamate) coated porous silica particles, 5 to 20 µm diameter	NUCLEOCEL *DELTA*	240

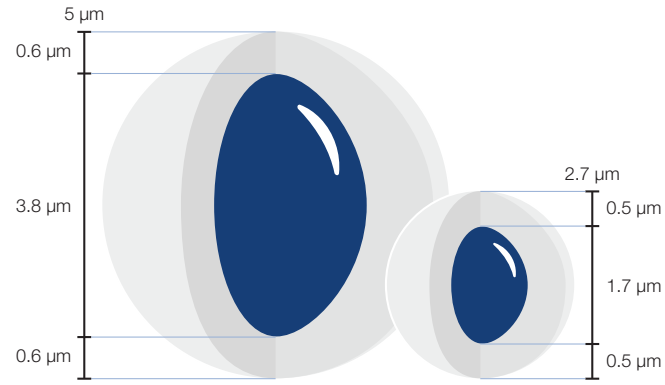
USP listing

USP specification of MN HPLC phases

Code	Specification	MN HPLC Phases	Page
USP L43	pentafluorophenyl groups chemically bonded to silica particles by a propyl spacer, 1.5 to 10 µm diameter	NUCLEODUR® PFP	174
		NUCLEOSHELL® PFP	212
USP L45	beta-cyclodextrin bonded to porous silica particles, R,S-hydroxypropyl ether derivative, 3 to 10 µm diameter	NUCLEODEX β-OH, β-PM	238
USP L58	strong cation-exchange resin consisting of sulfonated cross-linked PS/DVB copolymer in the Na form, 6 to 30 µm diameter	NUCLEOGEL® SUGAR Na	256
USP L60	spherical porous silica gel, particle size of 10 µm diameter or smaller, the surface of which has been covalently modified with alkyl amide groups and endcapped	NUCLEODUR® PolarTec	168
		NUCLEOSIL® C_{18} Nautilus	220
USP L75	A chiral-recognition protein, bovine serum albumin (BSA), chemically bonded to silica particles, about 7 µm in diameter, with a pore size of 300 Angstrom	RESOLVOSIL BSA-7	241
USP L118	Aqueous polymerized C_{18} groups on silica particles, 1.2 to 5 µm in diameter	NUCLEODUR® C_{18} PAH	234
		NUCLEOSIL® C_{18} PAH	236

NUCLEOSHELL® core-shell silica for HPLC

Core-shell technology

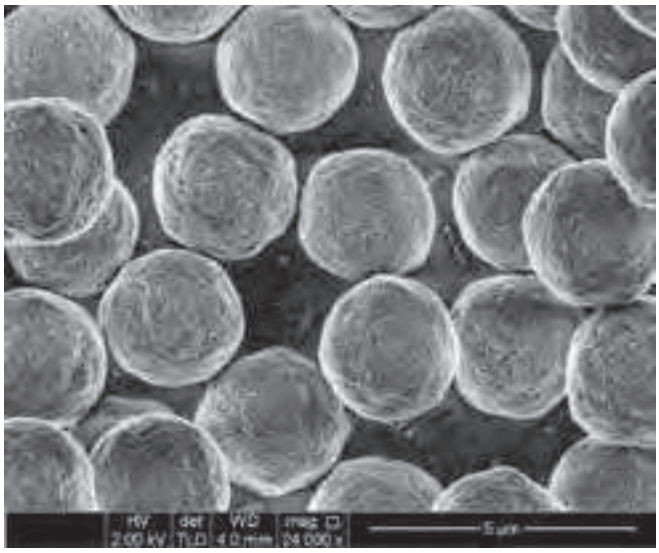


Key feature

- Solid core of silicon dioxide, homogeneous shell of porous silica
- Highest efficiency compared to traditional totally porous materials
- Pore size 90 Å; particle size 2.7 µm (core 1.7 µm) and 5 µm (core 3.8 µm); specific surface 130 (2.7 µm) and 90 (5 µm) m^2/g lower back pressure enables use on conventional LC systems
- Pressure stability 600 bar

Demands on HPLC separations are constantly increasing with respect to separation efficiency, detection limits, and the time requirements for each analysis.

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.



Electron microscopic image of NUCLEOSHELL®

NUCLEOSHELL® silica particles consist of a non-porous solid core of 1.7 µm diameter and a porous outer shell of 0.5 µm thickness. Accordingly the total diameter of the particle is 2.7 µm.

Utilizing a proprietary process of synthesis, NUCLEOSHELL® particles exhibit a distinct narrow particle size distribution (d_{90}/d_{10} ~ 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.

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

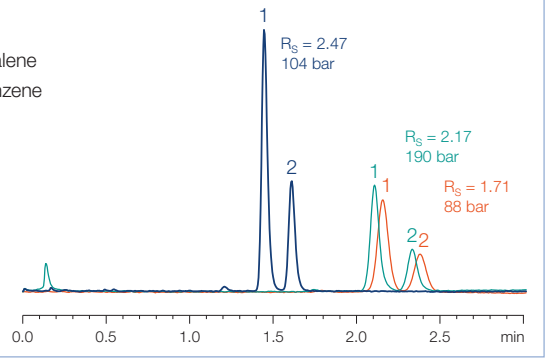
R_s = resolution, α = selectivity (separation factor), k_i' = retention
N = plate number with $N \propto 1/d_P$, d_P = particle diameter

Resolution R_S as function of particle size

MN Appl. No. 125270

Columns:	50 x 4 mm NUCLEOSHELL® RP 18, 2.7 µm NUCLEODUR® C_{18} Gravity, 3 µm NUCLEODUR® C_{18} Gravity, 1.8 µm
Eluent:	acetonitrile – water (60:40, v/v)
Flow rate:	1 mL/min
Temperature:	25 °C
Detection:	UV, 254 nm

Peaks:
1. Naphthalene
2. Ethylbenzene



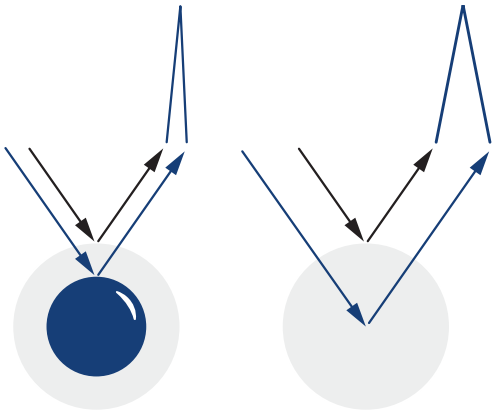
NUCLEOSHELL® core-shell silica for HPLC

Theoretical column efficiency (optimal conditions)

Silica	d_p [µm]	L [m]	HETP [µm]	Efficiency [plates/m]	L [mm]	N	R_s	Analysis time
NUCLEOSHELL®	2.7	1	4	250,000	100	25,000	112 %	40 %
	5	1	6.5	154,000	150	23,000	115 %	60 %
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

Core-shell particles vs. totally porous silica



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_{90}/d_{10} ~ 1.1)

- Stable packing

High heat transfer

- Minimized influence of frictional heat
- Efficiency of NUCLEOSHELL® ~ 250 000 m^{-1} (HETP ~ 4 µm)

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 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.

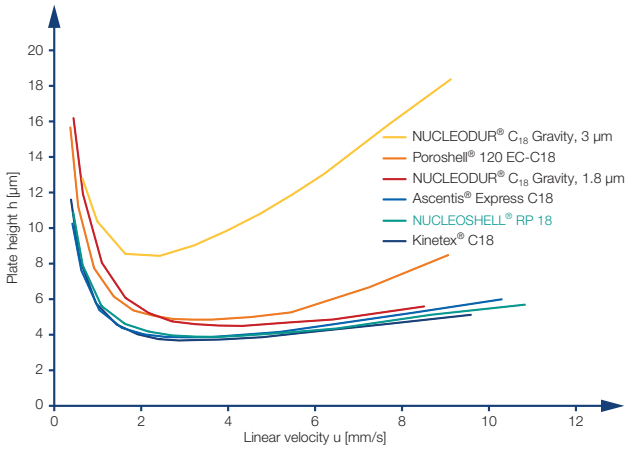
$$H = A + \frac{B}{u} + C \cdot u$$

A term = eddy-diffusion, B term = longitudinal diffusion coefficient, C term = mass transfer coefficient

Van Deemter curves

MN Appl. No. 125500

Column: 50 x 4.6 mm
Eluent: $CH_3CN - H_2O$ (70:30, v/v)
Temperature: 25 °C
Sample: acenaphthene



NUCLEOSHELL® core-shell silica for HPLC

In direct comparison with 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.

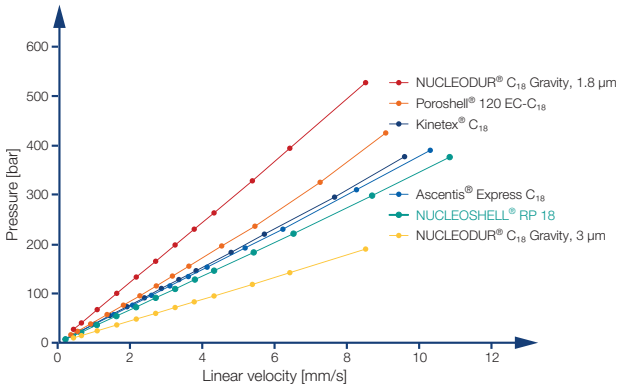
$$\Delta_P = \frac{\Phi \cdot L_C \cdot \eta \cdot u}{d_P^2}$$

Δ_P = pressure drop, Φ = flow resistance (non-dimensional), LC = column length, η = viscosity, u = linear velocity, d_P = particle diameter

Pressure drop

MN Appl. No. 125510

Column:	50 x 4.6 mm
Eluent:	CH_3CN – H_2O (70:30, v/v)
Temperature:	25 °C



Core-shell particle technology from MACHEREY-NAGEL is an alternate route to gain highest column efficiency and resolution in HPLC at short run time, but with moderate back pressure.

Features of NUCLEOSHELL® particles

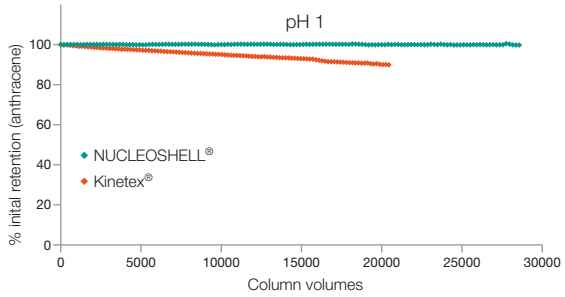
A criterion for the long-term stability of the column at pH extremes is the percentage decrease of initial retention and initial plates, respectively.

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

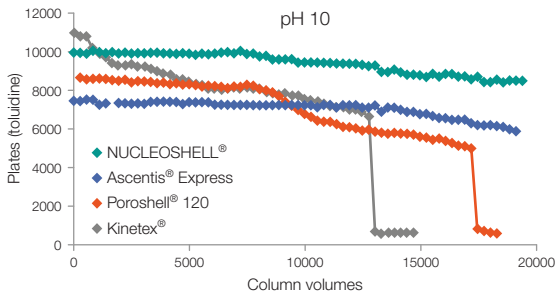
Stability under acidic and basic conditions

MN Appl. Nos. 125520 / 125530

Columns:	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 water, pH 1 (50:50, v/v)
Flow rate:	1.3 mL/min
Temperature:	80 °C
Detection:	UV, 254 nm
Analyt:	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/v)
Flow rate:	1.5 mL/min
Temperature:	40 °C
Detection:	UV, 220 nm
Analyt:	toluidine



Columns can be operated at elevated temperatures without loss in retention, efficiency or peak symmetry.

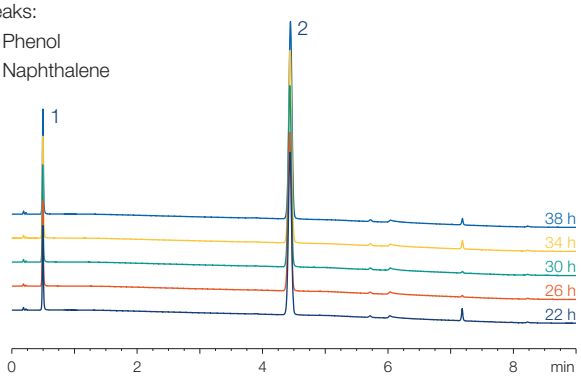
Temperature stability

MN Appl. No. 125400

Stability test:

Column:	50 × 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



Efficiency test:

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

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

NUCLEOSHELL® core-shell silica for HPLC

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

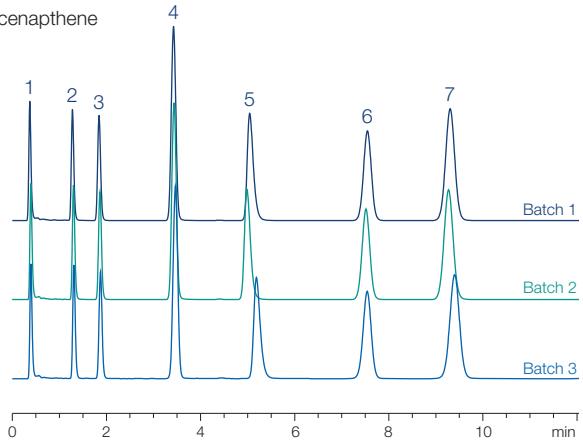
Batch-to-batch reproducibility

MN Appl. No. 125410

Column:	50 x 4 mm NUCLEOSHELL® RP 18, 2.7 µm
Eluent:	methanol – 25 mmol/L KH_2PO_4, pH 7 (70:30, v/v)
Flow rate:	1 mL/min
Temperature:	40 °C
Detection:	UV, 254 nm

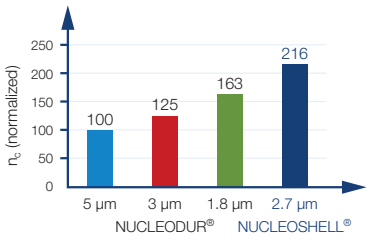
Peaks:

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



Peak capacity

The peak capacity is a measure for the number of sample analytes that can be separated on HPLC columns per time unit. Narrow peaks increase the peak capacity and thus the efficiency of the analytical column.



Comparison of peak capacities

$$n_c = 1 + \left(\frac{t_g}{W}\right)$$

n_c: peak capacity
t_g: gradient time
W: peak width (at baseline)

The example shows, that in comparison with totally porous NUCLEODUR® silica (1.8 µm) NUCLEOSHELL® provides 33 % higher peak capacity.

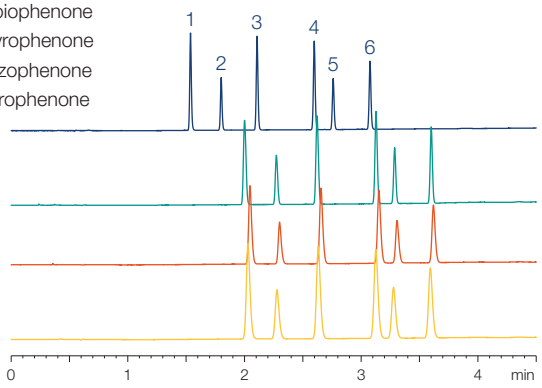
Peak capacity

MN Appl. No. 125540

Columns:	100 × 4.6 mm each NUCLEOSHELL® RP 18, 2.7 µm NUCLEODUR® C_{18} Gravity, 1.8 µm NUCLEODUR® C_{18} Gravity, 3 µm NUCLEODUR® C_{18} Gravity, 5 µm
Eluent:	A) acetonitrile, B) water, 40 – 100 % A in 4 min
Flow rate:	1.5 mL/min
Temperature:	25 °C
Detection:	UV, 230 nm

Peaks:

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

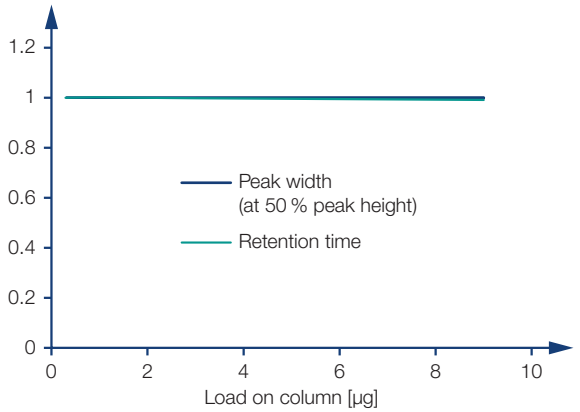


	Max. pressure [bar]	Resolution (4.5)
NUCLEOSHELL®, 2.7 µm	255	5.45
NUCLEODUR®, 1.8 µm	450	4.14
NUCLEODUR®, 3 µm	214	2.97
NUCLEODUR®, 5 µm	142	2.30

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 columns load although core-shell particles are suspected of showing a slightly lower loading capacity compared to fully porous silica materials.

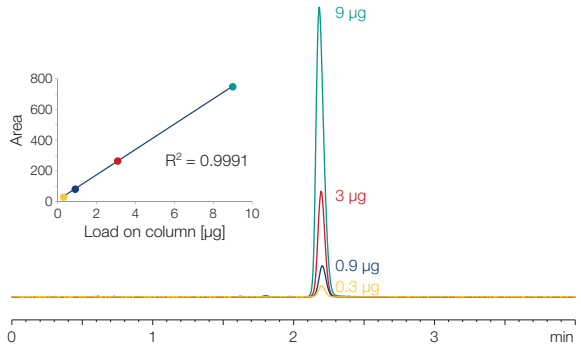
Normalized column parameters

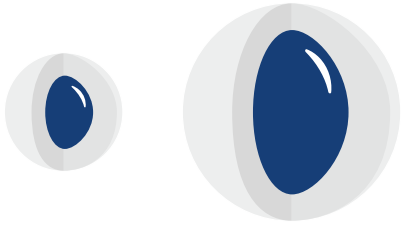


Loading capacity

Column: 50 x 3 mm NUCLEOSHELL® RP 18, 2.7 µm
Eluent: acetonitrile – 25 mmol/L KH_2PO_4, pH 3 (70:30, v/v)
Flow rate: 0.66 mL/min
Temperature: 30 °C
Detection: UV, 285 nm

Peaks:
1. Valerophenone





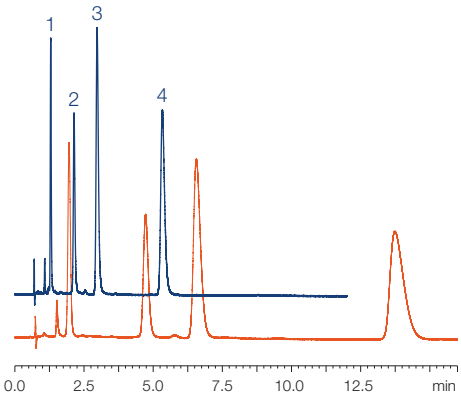
Method transfer of 5 µm particle columns

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.

Separation of cephalosporin antibiotics

MN Appl. No. 126630

Comparison of 5 µm core-shell and totally porous phase
Columns: each 100 x 4.6 mm
A) NUCLEOSHELL® RP 18plus, 5 µm
B) NUCLEODUR® Gravity C_{18}, 5 µm
Eluent: methanol – water + 0.1 % formic acid (35:65, v/v)
Flow rate: 1.3 mL/min
Pressure: 182 bar, 219 bar
Temperature: 25 °C
Detection: UV, 254 nm
Injection: 4.0 µL



Peaks:	Ret. time [min]		Asymmetry (EP)		Plates (EP)	
	A	B	A	B	A	B
1 Cefotaxime	1.30	1.96	1.19	1.12	6800	2218
2 Cefoxitin	2.14	4.72	1.22	1.20	6599	3471
3 Cefamandole	2.97	6.57	1.24	1.25	6259	3367
4 Cefalotine	5.33	13.73	1.32	1.61	6948	3672

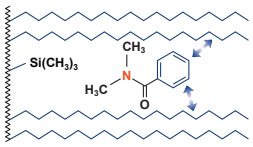
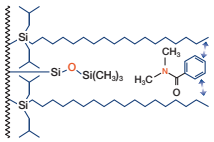
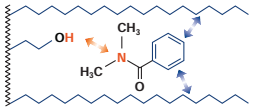
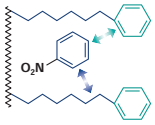
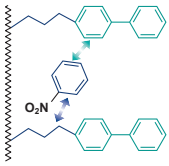
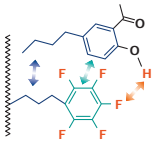
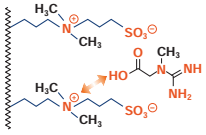
NUCLEOSHELL® phase overview

Overview of NUCLEOSHELL® HPLC phases

Phase	Specification	Page	Characteristic*	Stability	Structure
RP 18	octadecyl, multi-endcapping 7.8 % C (2.7 µm particles) 6.1 % C (5 µm particles) USP L1	200	A ●●●●● B ◖ C ●●◖	pH 1 – 11, suitable for LC/MS	NUCLEOSHELL® (Si-O$_2$)$_n$ NUCLEOSHELL® RP 18 –TMS
RP 18plus	octadecyl (monomeric), multi-endcapping 5.7 % C (2.7 µm particles) 4.4 % C (5 µm particles) USP L1	202	A ●●●● B ●●◖ C -	pH 2 – 9, suitable for LC/MS	NUCLEOSHELL® (Si-O$_2$)$_n$ NUCLEOSHELL® RP 18plus –TMS
Bluebird RP 18	octadecyl, hydrophilic endcapping 5 % C (2.7 µm particles) USP L1	204	A ●●●● B ●●◖ C ●●	stable in 100 % aqueous eluent, pH 1 – 8, suitable for LC/MS	NUCLEOSHELL® (Si-O$_2$)$_n$ NUCLEOSHELL® Bluebird RP 18 –OH –TMS –OH
Phenyl-Hexyl	phenylhexyl, multi-endcapping 4.5 % C (2.7 µm particles) USP L11	207	A ●● B ●●● C ●	pH 1 – 10, suitable for LC/MS	NUCLEOSHELL® (Si-O$_2$)$_n$ NUCLEOSHELL® Phenyl-Hexyl –TMS
Biphenyl	biphenylpropyl, multi-endcapping 5.2 % C (2.7 µm particles) USP L11	209	A ●●● B ●●● C ●●●	stable in 100 % aqueous eluent, pH 1.5 – 8.5, suitable for LC/MS	NUCLEOSHELL® (Si-O$_2$)$_n$ NUCLEOSHELL® Biphenyl –TMS
PFP	pentafluorophenyl, multi-endcapping ~ 3 % C (2.7 µm particles) USP L43	212	A ●● B ●●●● C ●●●●	pH 1 – 9, suitable for LC/MS	NUCLEOSHELL® (Si-O$_2$)$_n$ NUCLEOSHELL® PFP –TMS
HILIC	zwitterionic ammonium-sulfonic acid, no endcapping 1.3 % C (2.7 µm particles)	214	A ● B ●●●●● C -	pH 2 – 8.5, suitable for LC/MS	NUCLEOSHELL® (Si-O$_2$)$_n$ NUCLEOSHELL® HILIC –Si–OH

* A = ● hydrophobic selectivity, B = ● polar / ionic selectivity, C = ● steric selectivity
** phases which provide a similar selectivity based on chemical and physical properties

NUCLEOSHELL® phase overview

Application	Similar phases**	Interactions · retention mechanism	
overall sophisticated analytical separations, e. g., analgesics, anti-inflammatory drugs, antidepressants; herbicides; phytopharmaceuticals; immunosuppressants	Kinetex® C18; Cortecs® C18; Raptor® C18; Accucore® C18; Ascentis® Express C18; HALO® C18; Shim-pack Velox® C18	hydrophobic (van der Waals interactions)	
overall sophisticated analytical separations, especially for polar compounds, e. g., pharmaceuticals like antibiotics, water-soluble vitamins, organic acids	Kinetex® XB-C18; Bonshell® ASB-C18; Raptor® ARC-C18; Shim-pack Velox® SP-18	hydrophobic (van der Waals interactions)	
overall sophisticated analytical separations, especially for very polar compounds, e. g., pesticides, sweeteners, nitrosamines, water-soluble vitamins, organic acids, pharmaceuticals	Kinetex® Polar C_{18}	hydrophobic and polar (H bonds)	
aromatic and unsaturated compounds, polar compounds like pharmaceuticals, antibiotics	Ascentis® Express Phenyl-Hexyl; Kinetex® Phenyl-Hexyl; Accucore® Phenyl-Hexyl; Ultracore® Phenyl-Hexyl; Poroshell® Phenyl-Hexyl; HALO® Phenyl-Hexyl	π-π and hydrophobic	
aromatic and unsaturated compounds, mycotoxins, phthalates, hormones, polar compounds like pharmaceuticals, antibiotics, pesticides	Kinetex® Biphenyl, Raptor® Biphenyl, HALO® Biphenyl; Shim-pack Velox® Biphenyl	π-π and hydrophobic	
aromatic and unsaturated compounds, phenols, halogenated hydrocarbons, isomers, polar compounds like pharmaceuticals, antibiotics	Kinetex® PFP; Ascentis® Express F5; Accucore® PFP; Shim-pack Velox® PFP; HALO® PFP; Raptor® PFP	polar (H bond), dipole-dipole, π-π and hydrophobic	
hydrophilic compounds such as organic polar acids and bases, polar natural compounds	–	ionic / hydrophilic and electrostatic	

NUCLEOSHELL® columns

NUCLEOSHELL® RP 18 nonpolar high density phase · USP L1

Key feature

- Core-shell 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 data

- Octadecyl modification, multi-endcapped; pore size 90 Å, particle size 2.7 and 5 µm, carbon content 7.8 % for 2.7 µm, 6.1 % for 5 µm; pH stability 1 – 11; suitable for LC/MS

Recommended application

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

NUCLEOSHELL® RP 18 is based on core-shell silica. A unique derivatization process generates a homogeneous surface with a high density of bonded silanes. 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 below shows a sharp elution profile (superior resolution!) of these highly polar compounds with an excellent asymmetry value for amitriptyline of 1.12.

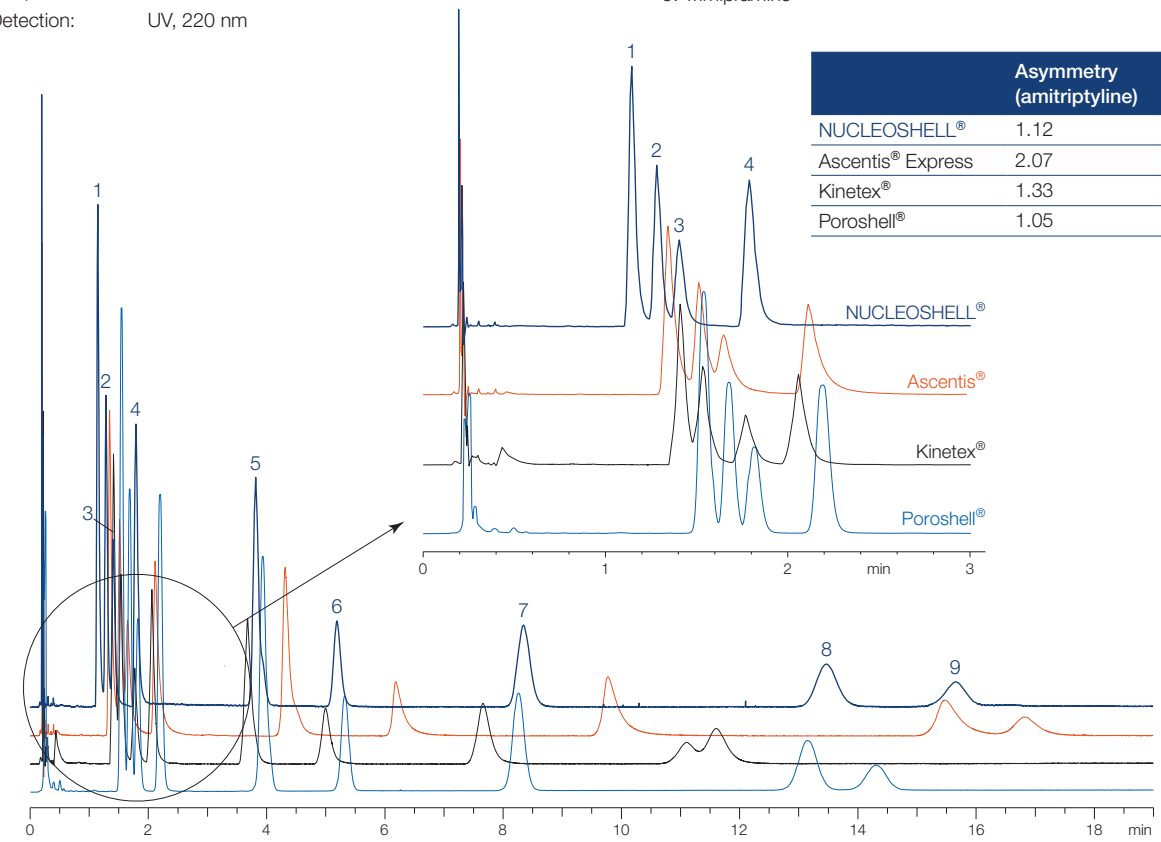
Tricyclic antidepressants · comparison of selectivity and resolution

MN Appl. No. 124960

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_2PO_4, 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

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

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



NUCLEOSHELL® columns

NUCLEOSHELL® RP 18 combines innovative silica technology and excellent surface deactivation, that outperforms conventional C_{18} 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 in many cases permits the use of existing HPLC equipment. 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 analyses in modern HPLC.

The separation of 13 β-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 β-lactam antibiotics in less than 3 min

MN Appl. No. 124940

Columns: 50 x 4 mm NUCLEOSHELL® RP 18, 2.7 µm
150 x 4 mm NUCLEODUR® C_{18} Gravity, 5 µm

Eluent: A) acetonitrile B) 20 mmol/L KH_2PO_4, 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

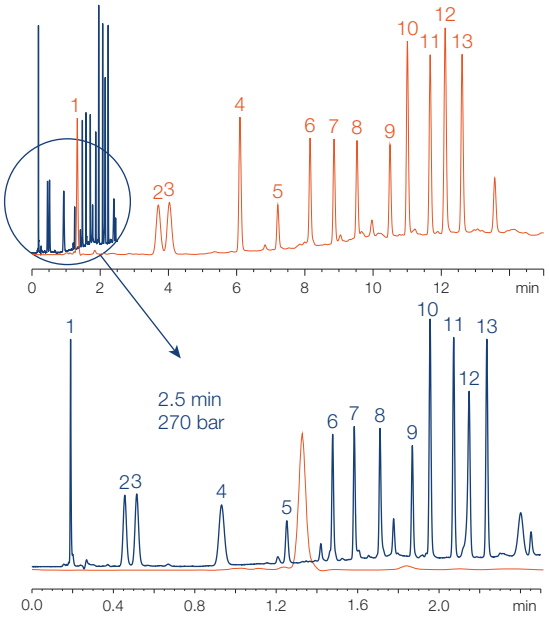
Pressure: 270 bar, 110 bar

Temperature: 25 °C

Detection: UV, 220 nm

Peaks:

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



Eluent in column acetonitrile – water

	ID	Length → 50 mm	100 mm	150 mm	250 mm	EC guard columns*
NUCLEOSHELL® RP 18, 2.7 µm; particle size 2.7 µm						
Analytical EC columns						
	2 mm	763132.20	763134.20	763136.20		763138.20
	3 mm	763132.30	763134.30	763136.30		763138.30
	4 mm	763132.40	763134.40	763136.40		763138.30
	4.6 mm	763132.46	763134.46	763136.46		763138.30
NUCLEOSHELL® RP 18, 5 µm; particle size 5 µm						
Analytical EC columns						
	2 mm	763152.20	763154.20	763156.20	763157.20	763158.20
	3 mm	763152.30	763154.30	763156.30	763157.30	763158.30
	4 mm	763152.40	763154.40	763156.40	763157.40	763158.30
	4.6 mm	763152.46	763154.46	763156.46	763157.46	763158.30

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

Guard column system

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

For details of the EC column system please see page 258.

NUCLEOSHELL® columns

NUCLEOSHELL® RP 18plus C_{18} phase with polar selectivity · USP L1

Key feature

- Based on core-shell particle technology for fast and efficient HPLC
- Hydrophobic C_{18} phase with distinct polar selectivity, ideal for method development
- Excellent performance under highly aqueous conditions

Technical data

- Monomeric octadecyl modification, multi-endcapped; 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

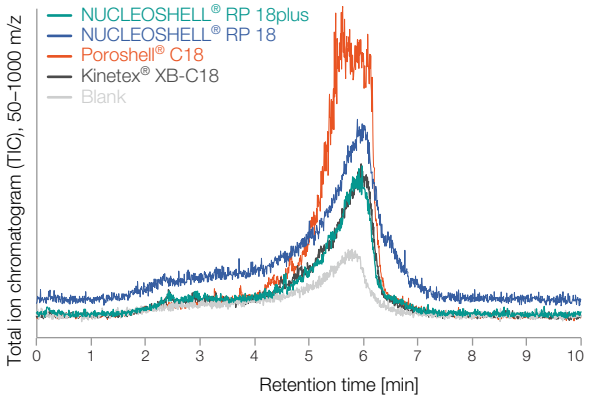
- Overall sophisticated analytical separations, especially for polar compounds, e. g., pharmaceuticals like antibiotics, water-soluble vitamins, organic acids

NUCLEOSHELL® RP 18plus is a C_{18} 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 with reduced steric selectivity compared to NUCLEOSHELL® RP 18.

Bleeding characterisitics

MN Appl. No. 126640

Column:	50 x 2 mm NUCLEOSHELL® RP 18plus, 2.7 µm
Eluent:	A) 0.1 % formic acid in water B) 0.1 % formic acid in acetonitrile 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

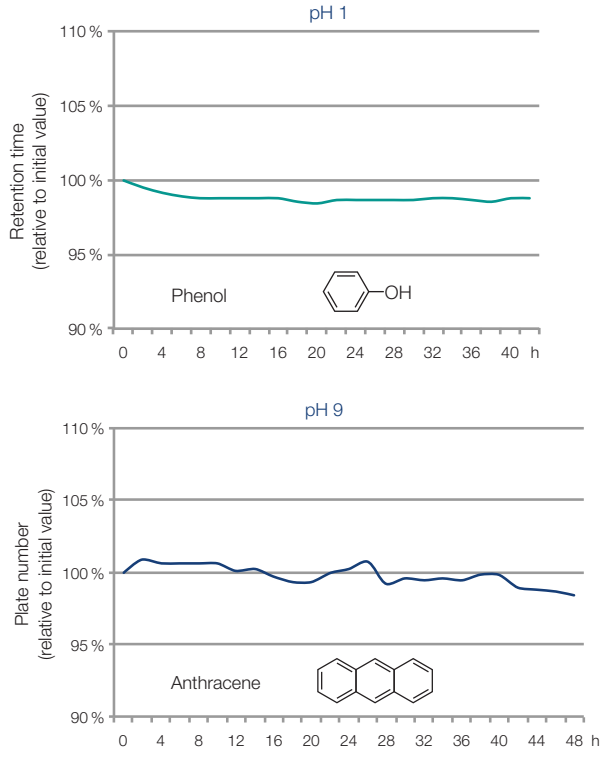


NUCLEOSHELL® RP 18plus combines superbly hydrophobic and polar selectivity – so it is a useful tool for method development in RP chromatography. Good pH stability and low bleeding characteristics make it ideal especially for LC/MS applications.

pH stability of NUCLEOSHELL® RP 18plus

MN Appl. No. 126650

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

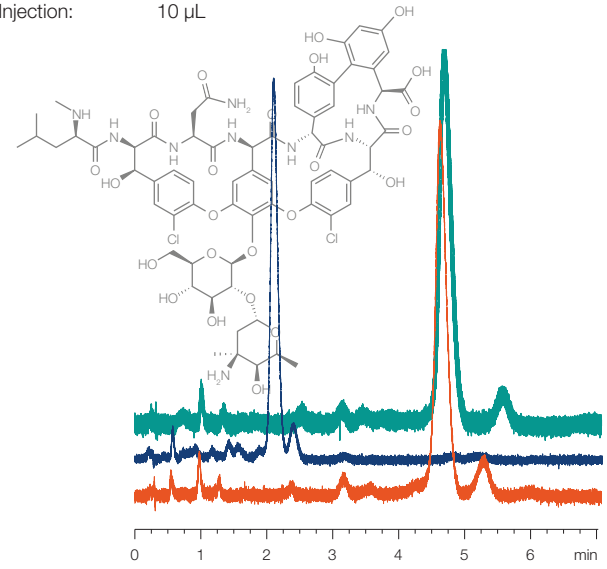


Also a comparison of retention of the glycopeptide antibiotic vancomycin on several octadecyl modified core-shell phases underlines the polar selectivity of NUCLEOSHELL® RP 18plus.

Polar selectivity shown for vancomycin

MN Appl. No. 126660

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

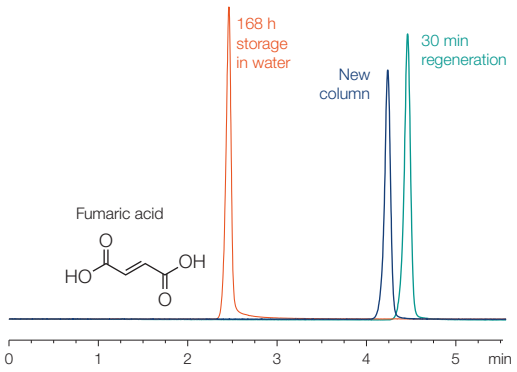


In addition NUCEOSHELL® RP 18plus provides a good stability under highly aqueous conditions. Even by long term usage or storage of the phase phase collapse and loss of retention are hardly observed. The original performance can be regained after a short regeneration procedure.

Phase collapse and regeneration

MN Appl. No. 126670

Column:	100 x 4 mm NUCLEOSHELL® RP 18plus, 2.7 µm
Eluent:	20 mmol/L KH_2PO_4, pH 2.6
Flow rate:	0.5 mL/min
Temperature:	20 °C
Detection:	UV, 215 nm
Injection:	0.5 µL



Eluent in column acetonitrile – water

	ID	Length → 50 mm	100 mm	150 mm	250 mm	EC guard columns*
NUCLEOSHELL® RP 18plus, 2.7 µm; particle size 2.7 µm						
Analytical EC columns						
	2 mm	763232.20	763234.20	763236.20		763238.20
	3 mm	763232.30	763234.30	763236.30		763238.30
	4 mm	763232.40	763234.40	763236.40		763238.30
	4.6 mm	763232.46	763234.46	763236.46		763238.30
NUCLEOSHELL® RP 18plus, 5 µm; particle size 5 µm						
Analytical EC columns						
	2 mm	763252.20	763254.20	763256.20	763257.20	763258.20
	3 mm	763252.30	763254.30	763256.30	763257.30	763258.30
	4 mm	763252.40	763254.40	763256.40	763257.40	763258.30
	4.6 mm	763252.46	763254.46	763256.46	763257.46	763258.30
EC columns in packs of 1, guard columns in packs of 3.						

Guard column system

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

For details of the EC column system please see page 258.

NUCLEOSHELL® columns

NUCLEOSHELL® Bluebird RP 18 for highly aqueous mobile phases · USP L1

Key feature

- Special core-shell phase with hydrophilic endcapping
- Stable in 100 % aqueous mobile phase
- Distinct polar selectivity features
- Excellent base deactivation; suitable for LC/MS due to low bleeding characteristics

Technical data

- Octadecyl phase; polar endcapped
- Pore size 90 Å; particle size 2.7 µm. carbon content 5 %; pH stability 1 – 8

Recommended application

- USP listing L1
- Pesticides, pharmaceuticals, water-soluble vitamins, sweeteners, nitrosamines, organic acids, very polar analytes

NUCLEOSHELL® Bluebird RP 18 is an octadecyl modified superficially porous silica. Due to an excellent base deactivation and a special hydrophilic endcapping procedure, NUCLEOSHELL® Bluebird RP 18 is extremely durable in 100 % aqueous mobile phase.

A robust bonding chemistry leads to low bleeding characteristics and therefor an excellent suitability for LC/MS applications.

The polar surface chemistry of NUCLEOSHELL® Bluebird RP 18 leads to retention characteristics distinctly different from conventional C_{18} phases. Sulfa drugs and various polar drug analytes can be very well separated as shown in the following applications (MN application numbers 128340 and 128390).

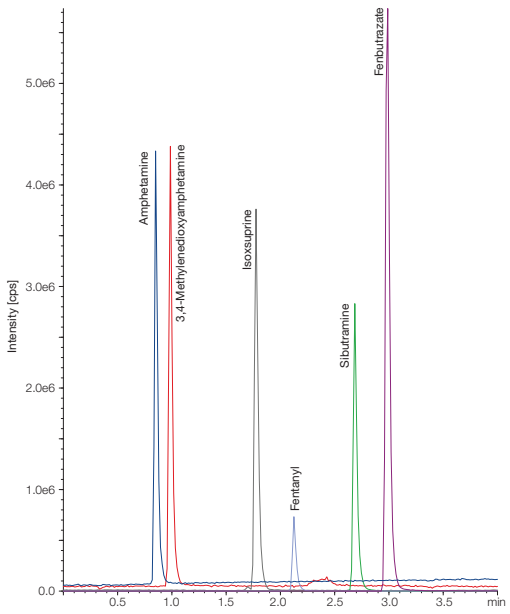
Drug analytes

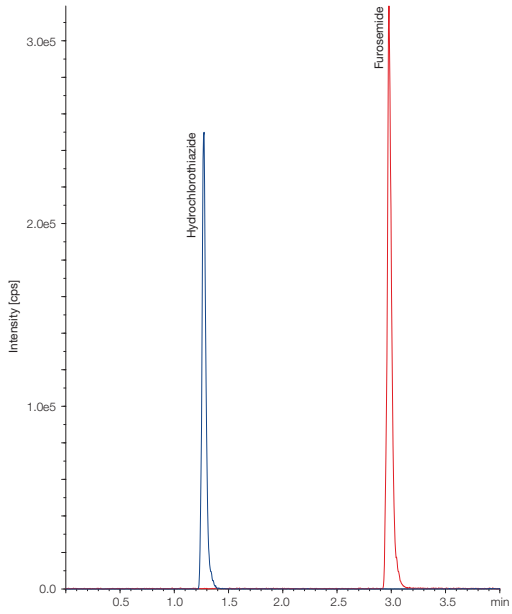
MN Appl. No. 128340

Column:	50 x 4.6 mm NUCLEOSHELL® Bluebird RP 18, 2.7 µm
Eluent:	A) 0.1 % formic acid in water B) 0.1 % formic acid in methanol
Gradient:	in 4.5 min from 5 % to 90 % B, hold for 0.5 min, in 0.5 min to 5 % B, hold 0 % B for 4.5 min
Flow rate:	1.3 mL/min
Temperature:	30 °C
Detection:	MS, SMRM
Injection:	5 µL
Concentration:	50 ng/mL for each analyte

MRM transitions

Analyte	RT [min]	$[M+H]^+$	Q_1 (Quantifier)	Q_2 (Qualifier)
Amphetamine	0.85	136.0	91.1	108.9
3,4-Methylenedi-oxyamphetamine	0.99	180.0	163.1	105.0
Isoxsuprine	1.78	303.0	285.1	77.1
Fentanyl	2.13	337.0	304.9	105.1
Sibutramine	2.69	280.0	125.0	139.1
Fenbutrazate	2.99	368.2	191.1	91.1
Analyte	**RT [min]**	**$[M-H]^-$**	**Q_1 (Quantifier)**	**Q_2 (Qualifier)**
Hydrochlorothiazide	1.27	295.9	268.7	98.9
Furosemide	2.98	329.0	283.2	255.2





Sulfa drugs

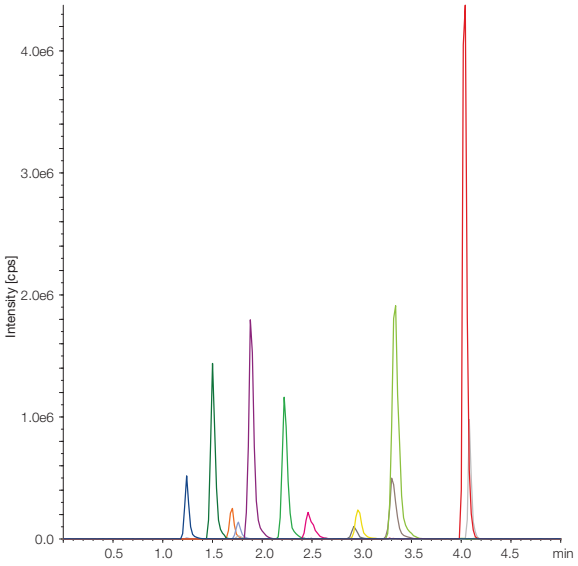
MN Appl. No. 128390

Column: 50 x 4.6 mm NUCLEOSHELL® Bluebird RP 18, 2.7 µm
Eluent: A) 0.1 % formic acid in water
B) 0.1 % formic acid in methanol
Gradient: in 4.0 min from 5 % to 20 % B, in 1.0 min to 80 % B, hold 80 % B for 0.5 min, in 0.1 min to 5 % B, hold 5 % B for 4.4 min
Flow rate: 1.3 mL/min
Temperature: 50 °C
Detection: MS, MRM
Injection: 5 µL
Concentration: 100 ng/mL for each analyte

MRM transitions

Analyte	RT [min]	$[M+H]^+$	Q_1 (Quantifier)	Q_2 (Qualifier)
Sulfacetamide	1.24	215.2	156.2	92.1
Sulfadiazine	1.50	251.2	156.1	92.1
Sulfapyridine	1.69	250.2	156.1	92.0
Sulfatiazole	1.75	256.2	156.2	92.1
Sulfamerazine	1.89	265.1	156.1	92.1
Sulfadimidine	2.22	279.2	185.9	65.0
Sulfamethoxypyridazine	2.46	281.2	156.1	92.2
Sulfamonomethoxine	2.92	281.2	156.1	92.2
Sulfachlorpyridazine	2.96	285.2	156.1	92.1
Sulfamethoxazole	3.31	254.2	156.1	92.1
Sulfadoxine	3.72	311.1	156.1	92.1

Analyte	RT [min]	$[M+H]^+$	Q_1 (Quantifier)	Q_2 (Qualifier)
Sulfadimethoxine	4.03	311.1	156.1	92.1
Sulfaquinoxaline	4.08	301.2	156.1	92.1



The reliable phase modification process leads to a high batch-to-batch reproducibility, where different batches show very consistent performance results. This can be shown in application 128610 with analytes of different polarities, which also demonstrate the hydrophobic properties of this C_{18} phase.

Batch-to-batch reproducibility

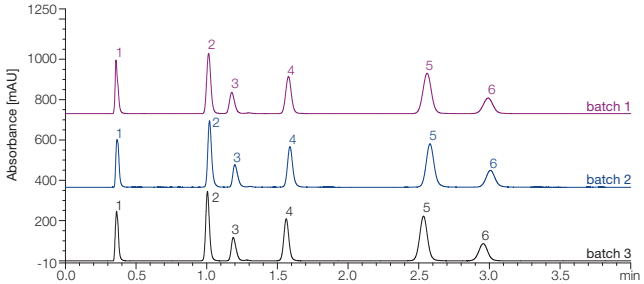
MN Appl. No. 128610

Column: 50 x 4 mm NUCLEOSHELL® Bluebird RP 18, 2.7 µm
Eluent: 25 mM ammonium dihydrogen phosphate solution – methanol (35:65, v/v), pH = 7.0
Flow rate: 1.0 mL/min
Temperature: 40 °C
Detection: UV, 254 nm
Injection: 5 µL
Concentration:

Uracil	45 µg/mL
Ethyl benzoate	181 µg/mL
Lidocaine	1134 µg/mL
Naphthalene	1134 µg/mL
Biphenyl	45 µg/mL
Acenaphthene	227 µg/mL

The mixture was diluted to 4 mL with water

Peaks:
1. Uracil
2. Ethyl benzoate
3. Lidocaine
4. Naphthalene
5. Biphenyl
6. Acenaphthene



NUCLEOSHELL® columns

In addition even very polar organic acids can be analyzed while retaining an excellent performance on NUCLEOSHELL® Bluebird RP 18 using 100 % aqueous mobile phase.

Organic acids

MN Appl. No. 128330

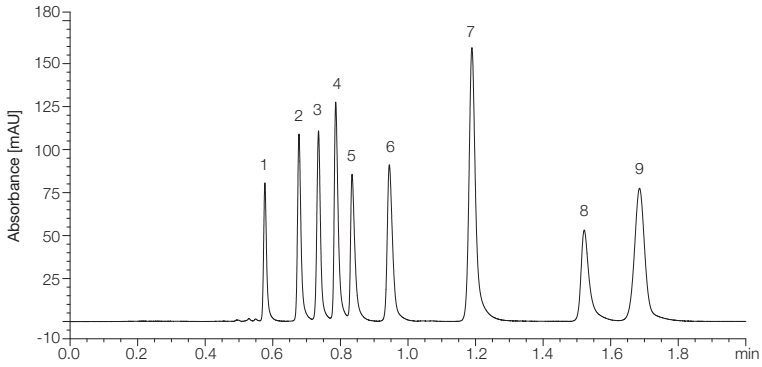
Column:	150 x 4 mm NUCLEOSHELL® Bluebird RP 18, 2.7 µm
Eluent:	50 mM potassium dihydrogen phosphate solution, pH = 2.5
Flow rate:	2.0 mL/min
Temperature:	40 °C
Detection:	UV, 210 nm
Injection:	3 µL

Concentration (in water)

Tartaric acid	135 µg/mL
Malic acid	2162 µg/mL
Shikimic acid	27 µg/mL
Lactic acid	2703 µg/mL
Acetic acid	2703 µg/mL
Citric acid	1081 µg/mL
Fumaric acid	41 µg/mL
Acrylic acid	676 µg/mL
Arbutin	216 µg/mL

Peaks:

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



Length (mm)	ID (mm)	Particle size (µm)	REF	Guard columns*
NUCLEOSHELL® Bluebird RP 18 (pack of 1)				
Analytical EC columns				
150	4.6	2.7	763436.46	763438.30
150	4	2.7	763436.40	763438.30
150	3	2.7	763436.30	763438.30
150	2	2.7	763436.20	763438.20
100	4.6	2.7	763434.46	763438.30
100	4	2.7	763434.40	763438.30
100	3	2.7	763434.30	763438.30
100	2	2.7	763434.20	763438.20
50	4.6	2.7	763432.46	763438.30
50	3	2.7	763432.30	763438.30

* Pack of 3, EC guard columns require column protection system REF 718966. For more information, 259

NUCLEOSHELL® Phenyl-Hexyl Alternative selectivity to C_{18} phases · USP L11

Key feature

- Based on core-shell particle technology for fast and efficient HPLC
- Hydrophobic phase with alternative selectivity compared to classical C_{18} modifications
- Separation principle based on 2 retention mechanisms: π-π interactions and hydrophobic interactions

Technical data

- Phenyl-Hexyl modification, multi-endcapped; 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

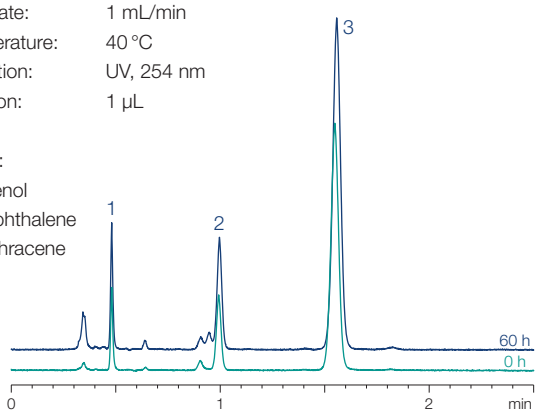
Phenyl-Hexyl modified phases offer an excellent separation efficiency especially for aromatic and unsaturated compounds with electron-withdrawing groups. The combination of hydrophobic and π-π interactions results in an alternative and interesting selectivity profile compared to C_{18} or C_8 modifications. NUCLEOSHELL® Phenyl-Hexyl is based on a 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.

Stability of NUCLEOSHELL® Phenyl-Hexyl at pH 10

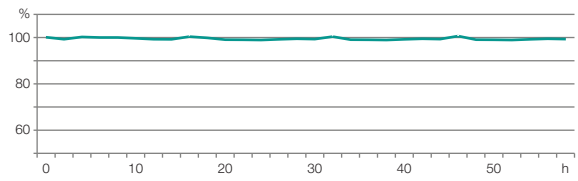
MN Appl. No. 126420

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

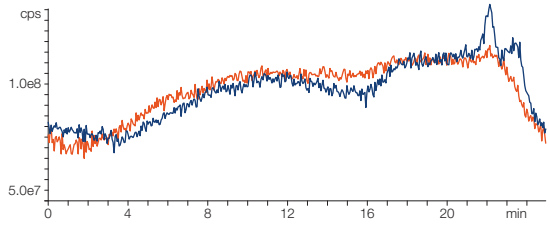


NUCLEOSHELL® Phenyl-Hexyl is a robust phase with an alternative RP selectivity for aromatic and unsaturated analytes compared to classical C_{18} / C_8 phases – it is an additional and useful tool for all chromatography users.

Bleeding characteristics of NUCLEOSHELL® Phenyl-Hexyl

MN Appl. No. 126400

Columns:	50 × 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



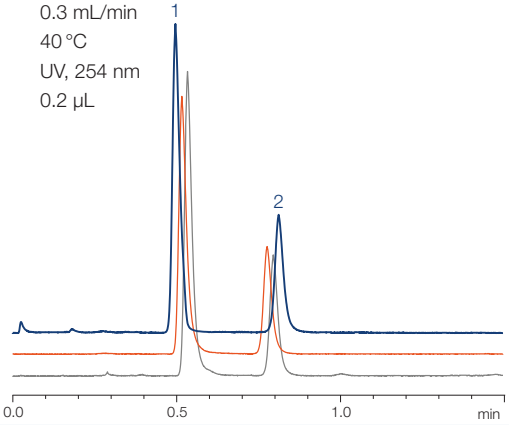
The pyridine-phenol test shows that NUCLEOSHELL® Phenyl-Hexyl provides a symmetrical peak for pyridine and higher resolution in comparison to other core-shell based Phenyl-Hexyl phases, which underlines the excellent base deactivation.

Pyridine-phenol test of NUCLEOSHELL® Phenyl-Hexyl

MN Appl. No. 126410

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
Injection:	0.2 µL

Peaks:
1. Pyridine
2. Phenol



NUCLEOSHELL® columns

Comparing the separation of sulfonamides on NUCLEODUR® Phenyl-Hexyl with different particle sizes

MN Appl. No. 125860

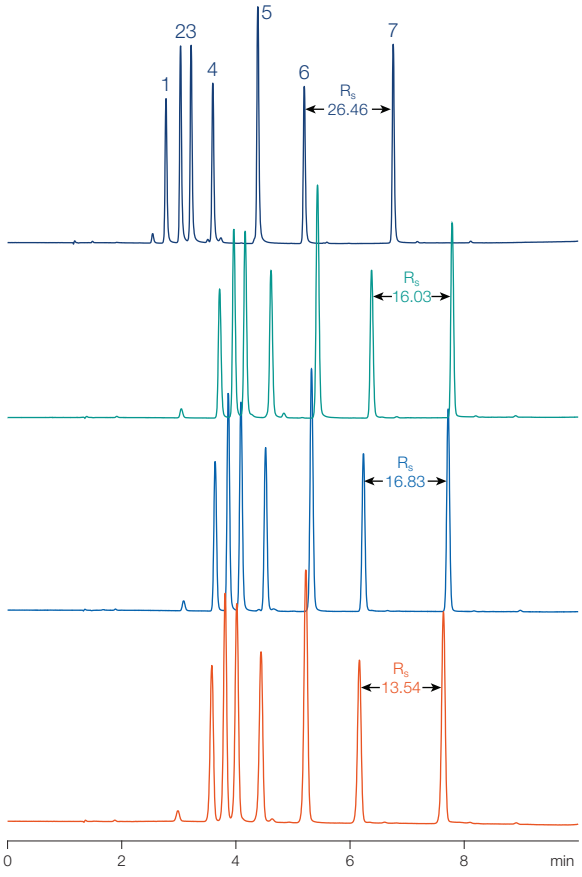
Columns: 150 × 3 mm each
NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm
NUCLEODUR® Phenyl-Hexyl, 1.8 µm
NUCLEODUR® Phenyl-Hexyl, 3 µm
NUCLEODUR® Phenyl-Hexyl, 5 µm

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. Sulfachlorpyridazine
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.



The separation of sulfonamides proves the scalability from fully porous NUCLEODUR® to NUCLEOSHELL® Phenyl-Hexyl. Hereby the core-shell silica exhibits identical selectivity, narrower peaks and slightly shorter retention under the same conditions.

Thus, method transferability between NUCLEODUR® and NUCLEOSHELL® is guaranteed, either for speeding up your methods or scaling up for preparative requirements.

Eluent in column acetonitrile – water

ID	Length → 50 mm	100 mm	150 mm	EC guard columns*
NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm; particle size 2.7 µm				
Analytical EC columns				
2 mm	763732.20	763734.20	763736.20	763738.20
3 mm	763732.30	763734.30	763736.30	763738.30
4 mm	763732.40	763734.40	763736.40	763738.30
4.6 mm	763732.46	763734.46	763736.46	763738.30
EC columns in packs of 1, guard columns in packs of 3.				

Guard column system

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

For details of the EC column system please see page 258.

NUCLEOSHELL® Biphenyl for highly aqueous mobile phases · USP L11

Key feature

- Enhanced retention for aromatic and unsaturated substances due to a separation principle based on 2 retention mechanisms: π-π interactions and hydrophobic interactions
- Stable in 100 % aqueous mobile phase systems
- Suitable for LC/MS due to low bleeding characteristics

Technical data

- Biphenylpropyl phase; multi-endcapped
- Pore size 90 Å; particle size 2.7 µm. carbon content 5.2 %; pH stability 1.5 – 8.5

Recommended application

- USP listing L11
- Pesticides, pharmaceuticals, mycotoxins, phthalates, hormones, DNPH aldehydes, aromatic and unsaturated compounds

NUCLEOSHELL® Biphenyl is a biphenyl modified superficially porous silica.

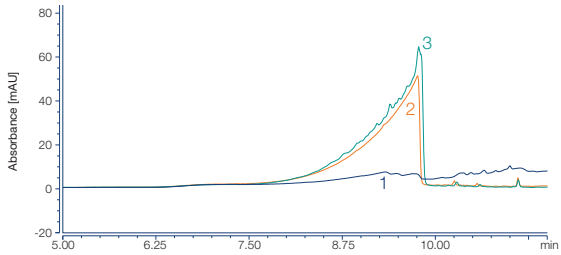
The special phase modification of NUCLEOSHELL® Biphenyl with iso-butyl sidechains leads to low bleeding characteristics even at very acidic pH values compared to competitor columns (as shown in application 128780). Due to these iso-butyl sidechains and multi-endcapping procedures no phase collapse occurs and stability in 100 % aqueous mobile phase is ensured. Additionally NUCLEOSHELL® Biphenyl shows an excellent suitability for LC/MS applications.

A reliable phase modification process guarantees a high batch-to-batch reproducibility. This can be shown in application 128760 with different analytes. The separation of these compounds with various polarities demonstrates the hydrophobic as well as polar properties of this biphenyl phase.

Stability in acidic medium (gradient method)

MN Appl. No. 128780

Column:	100 x 3 mm NUCLEOSHELL® Biphenyl, 2.7µm
Eluent:	A) 1 % H_3PO_4 (pH = 1.2) B) acetonitrile
Gradient:	equilibration 10 min 10 % B, hold 10 % B for 5 min, from 10 % to 90 % B in 5 min, hold 90 % B for 3 min, in 1.0 min to 10 % B
Flow rate:	0.56 mL/min
Temperature:	40 °C
Detection:	UV, 254 nm
1.	NUCLEOSHELL® Biphenyl, 2.7 µm
2.	Kinetex® Biphenyl, 2.6 µm
3.	Raptor® Biphenyl, 2.7 µm



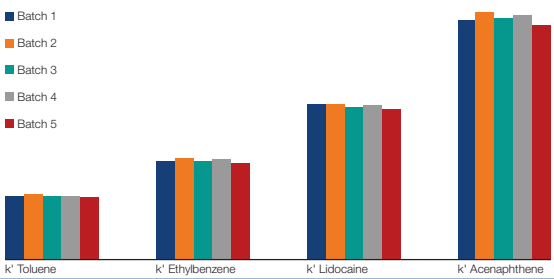
Batch-to-batch reproducibility

MN Appl. No. 128760

Column:	50 x 4 mm NUCLEOSHELL® Biphenyl, 2.7 µm
Eluent:	25 mM potassium dihydrogen phosphate solution – methanol (70:30, v/v), pH = 7.0
Flow rate:	1.0 mL/min
Run time:	10 min
Temperature:	30 °C
Detection:	UV, 254 nm
Injection:	1 µL

Concentration (in methanol)

Uracil	40 µg/mL (void volume marker)
Toluene	1250 µg/mL
Ethylbenzene	1250 µg/mL
Lidocaine	500 µg/mL
Acenaphthene	230 µg/mL



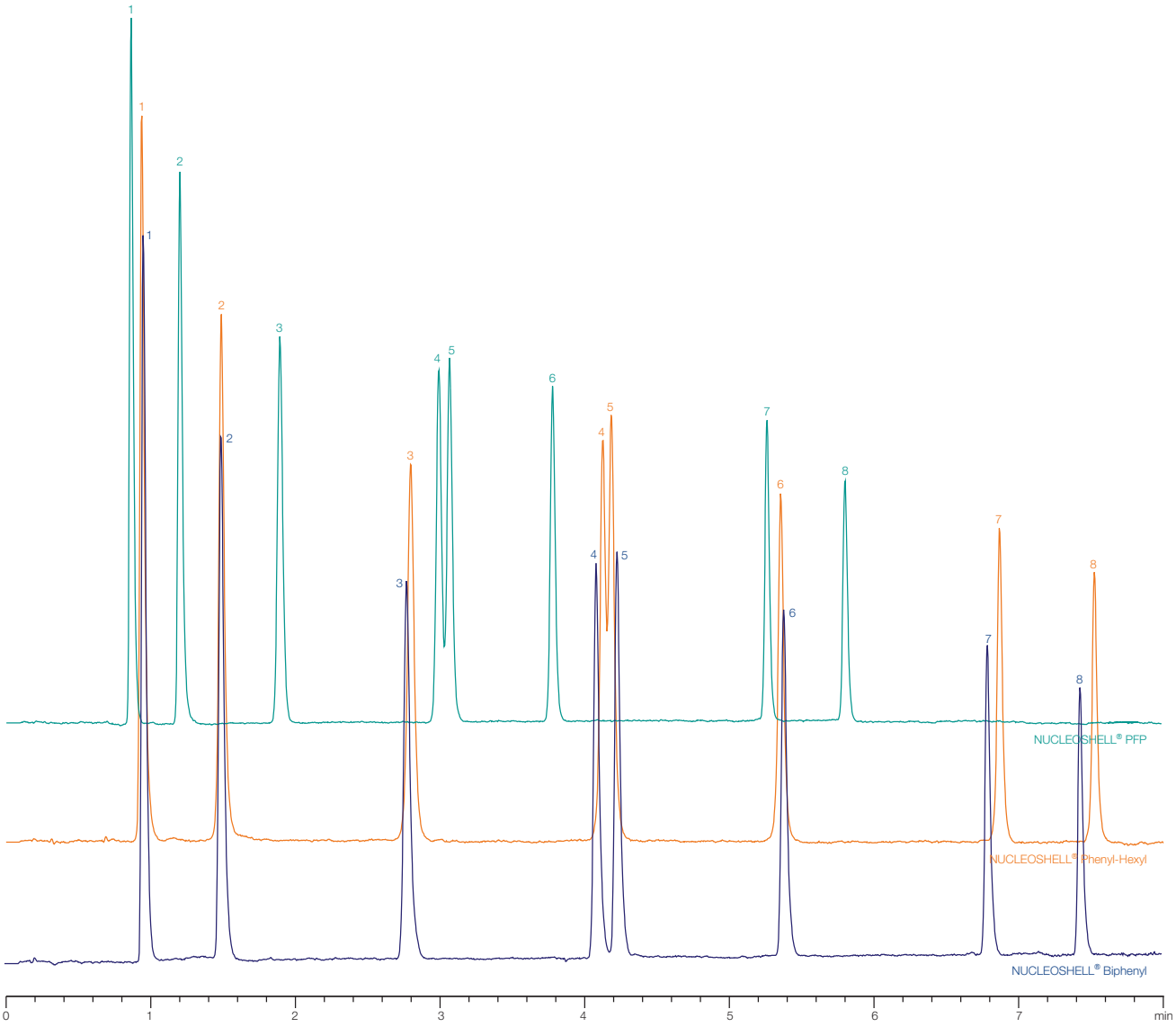
Phthalates

MN Appl. No. 128830

Columns:	100 x 3 NUCLEOSHELL® Biphenyl, 2.7 µm 100 x 3 NUCLEOSHELL® Phenyl-Hexyl, 2.7 µm 100 x 3 NUCLEOSHELL® PFP, 2.7 µm
Eluent:	A) water B) 0.1 % water in acetonitrile
Gradient:	hold 50 % B for 1.5 min, in 6.0 min to 95 % B, hold 95 % B for 3.5 min, in 2.0 min to 50 % B, hold 50 % B for 4.5 min
Flow rate:	1.0 mL/min
Temperature:	30 °C
Detection:	UV, 228 nm
Injection:	5 µL
Concentration:	10.0 ng/mL for each analyte in water – acetonitrile (1:1, v/v)

Retention times

	Analyte	Biphenyl RT [min]	Phenyl-Hexyl RT [min]	PFP RT [min]
1	Dimethyl phthalate	0.96	0.94	0.86
2	Diethyl phthalate	1.50	1.49	1.20
3	Dipropyl phthalate	2.87	2.80	1.89
4	Dibutyl phthalate	4.09	4.13	2.99
5	Benzyl butyl phthalate	4.24	4.19	3.07
6	Dicyclohexyl phthalate	5.39	5.36	3.78
7	Diheptyl phthalate	6.80	6.87	5.26
8	Dioctyl phthalate	7.44	7.53	5.80



Compared to other aryl HPLC modifications NUCLEOSHELL® Biphenyl shows more pronounced π-π interactions. In application 128830 NUCLEOSHELL® Biphenyl is able to separate the critical analyte pair dibutyl phthalate and benzyl butyl phthalate whereas other aryl phases cannot achieve a baseline separation.

Length (mm)	ID (mm)	Particle size (µm)	REF	Guard columns*
NUCLEOSHELL® Biphenyl (pack of 1)				
Analytical EC columns				
150	4.6	2.7	763636.46	763638.30
150	4	2.7	763636.40	763638.30
150	3	2.7	763636.30	763638.30
150	2	2.7	763636.20	763638.20
100	4.6	2.7	763634.46	763638.30
100	4	2.7	763634.40	763638.30
100	3	2.7	763634.30	763638.30
100	2	2.7	763634.20	763638.20
50	3	2.7	763632.30	763638.30
50	2	2.7	763632.20	763638.20

* Pack of 3, EC guard columns require column protection system REF 718966. For more information, see page 259.

NUCLEOSHELL® columns

NUCLEOSHELL® PFP hydrophobic pentafluorophenyl phase · USP L43

Key feature

- Core-shell technology for fast and efficient HPLC
- Hydrophobic phase with alternative selectivity in comparison to classical C_{18} modifications
- Separation principle based on 4 retention mechanisms (polar interactions (H bonds), dipole-dipole, π-π, hydrophobic interactions)

Technical data

- Phase with pentafluorophenylpropyl modification, multi-endcapping; 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, halogen compounds, isomers, polar compounds like pharmaceuticals, antibiotics; strong retention of basic compounds

Orthogonality in selectivity

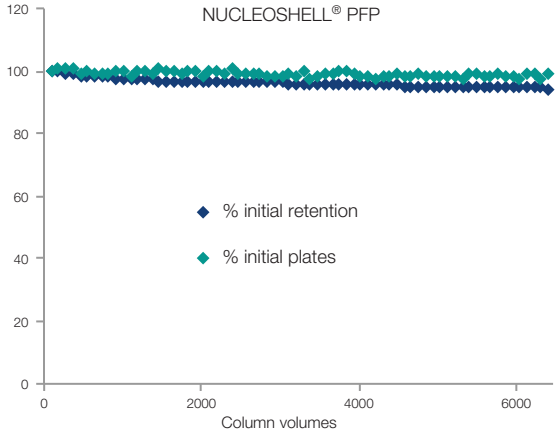
Fluorinated stationary phases in HPLC have gained increasing interest over the last years. Most common representative of fluorinated silica phases is the pentafluorophenyl modification (PFP or F_5). Especially the orthogonal selectivity compared to traditional alkyl phases widens the scope in analytical HPLC. Thus NUCLEOSHELL® PFP offers an excellent selectivity especially for highly polar analytes, aromatic and unsaturated compounds, phenols or halogenated hydrocarbons.

While a typical C_{18} phase just provides hydrophobic interactions between stationary phase and analyte NUCLEOSHELL® 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.

Stability of NUCLEOSHELL® PFP at pH 1

MN Appl. No. 125560

Columns:	100 x 4.6 mm NUCLEOSHELL® PFP, 2.7 µm 100 x 4.6 mm Kinetex® PFP, 2.6 µm F5
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



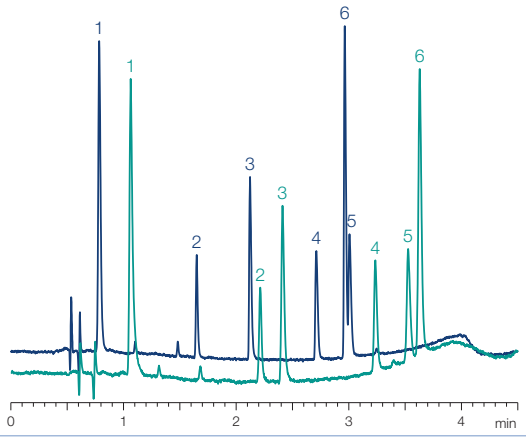
β-Blockers · orthogonal selectivity of NUCLEOSHELL® PFP

MN Appl. No. 125610

Columns:	100 × 4.6 mm NUCLEOSHELL® RP 18, 2.7 µm NUCLEOSHELL® PFP, 2.7 µm
Eluent:	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



Methylacetophenones

MN Appl. No. 125590

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 F5

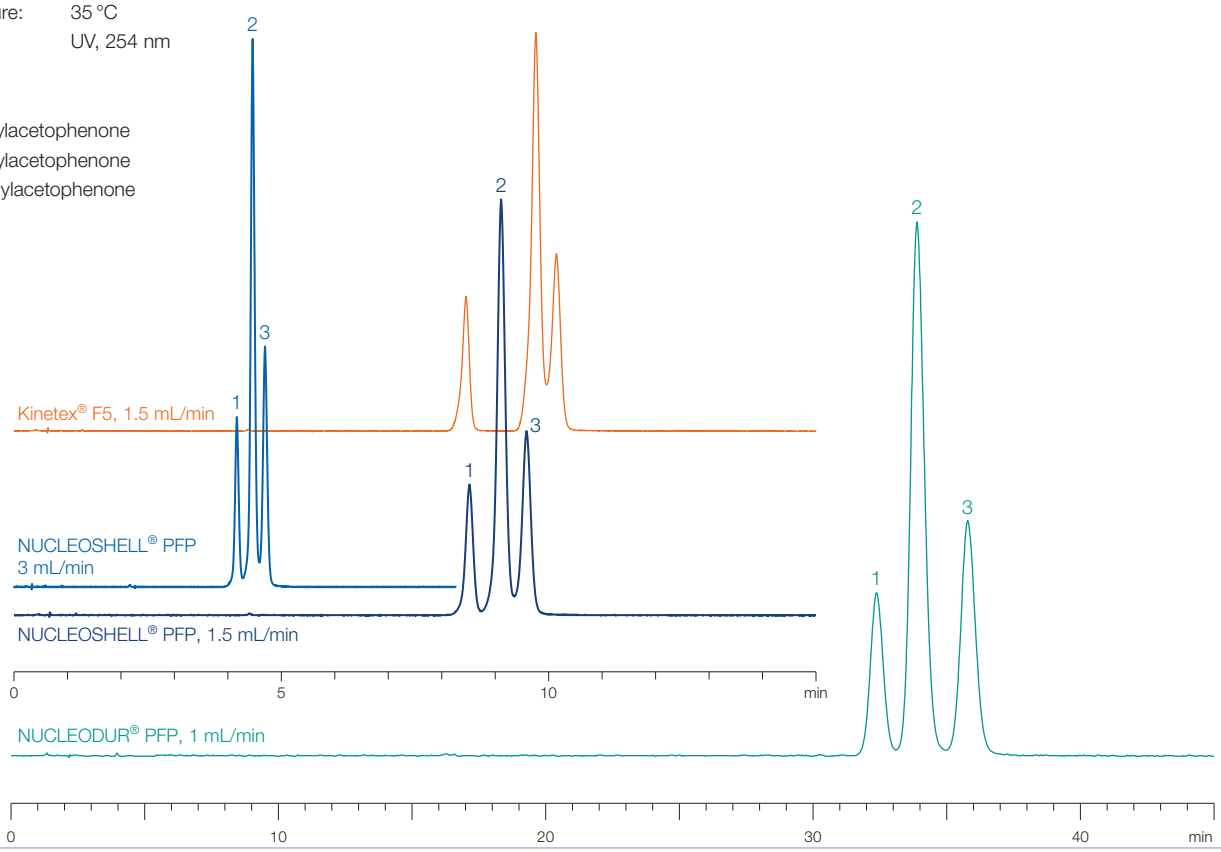
Eluent: Methanol – water (35:65, v/v)

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

Eluent in column acetonitrile – water

ID	Length → 50 mm	100 mm	150 mm	EC guard columns*
NUCLEOSHELL® PFP, 2.7 µm; particle size 2.7 µm				
Analytical EC columns				
2 mm	763532.20	763534.20	763536.20	763538.20
3 mm	763532.30	763534.30	763536.30	763538.30
4 mm	763532.40	763534.40	763536.40	763538.30
4.6 mm	763532.46	763534.46	763536.46	763538.30
EC columns in packs of 1, guard columns in packs of 3.				

Guard column system

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

For details of the EC column system please see page 258.

NUCLEOSHELL® columns

NUCLEOSHELL® HILIC zwitterionic phase

Key feature

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

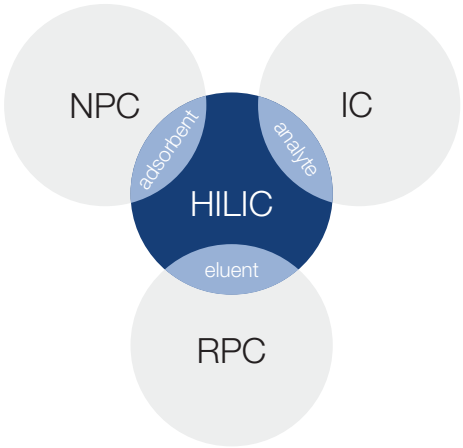
Technical data

- 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 polar organic acids and bases, polar natural compounds, nucleosides, oligonucleotides, amino acids, peptides, water-soluble vitamins

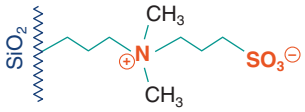
Hydrophilic interaction chromatography



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_8 or C_{18} 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. p nd.). The betaine character of the strong ion-exchanger results in full charge balancing and facilitates fast equilibration times.



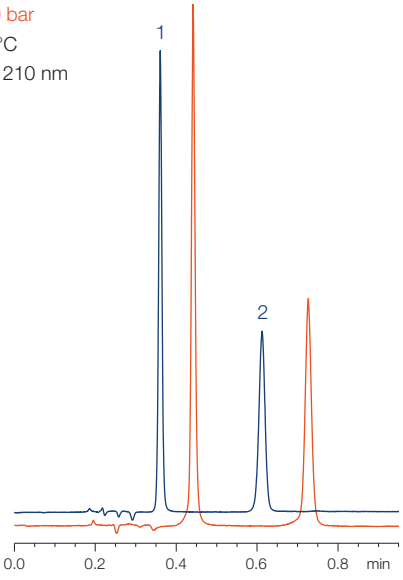
Good separation of polar compounds like the physiologically important substances creatine and creatinine can be achieved on NUCLEOSHELL® HILIC as well as on NUCLEODUR® HILIC, 1.8 µm at similar retention, but much lower back pressure.

Separation of creatine and creatinine

MN Appl. No. 124990

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

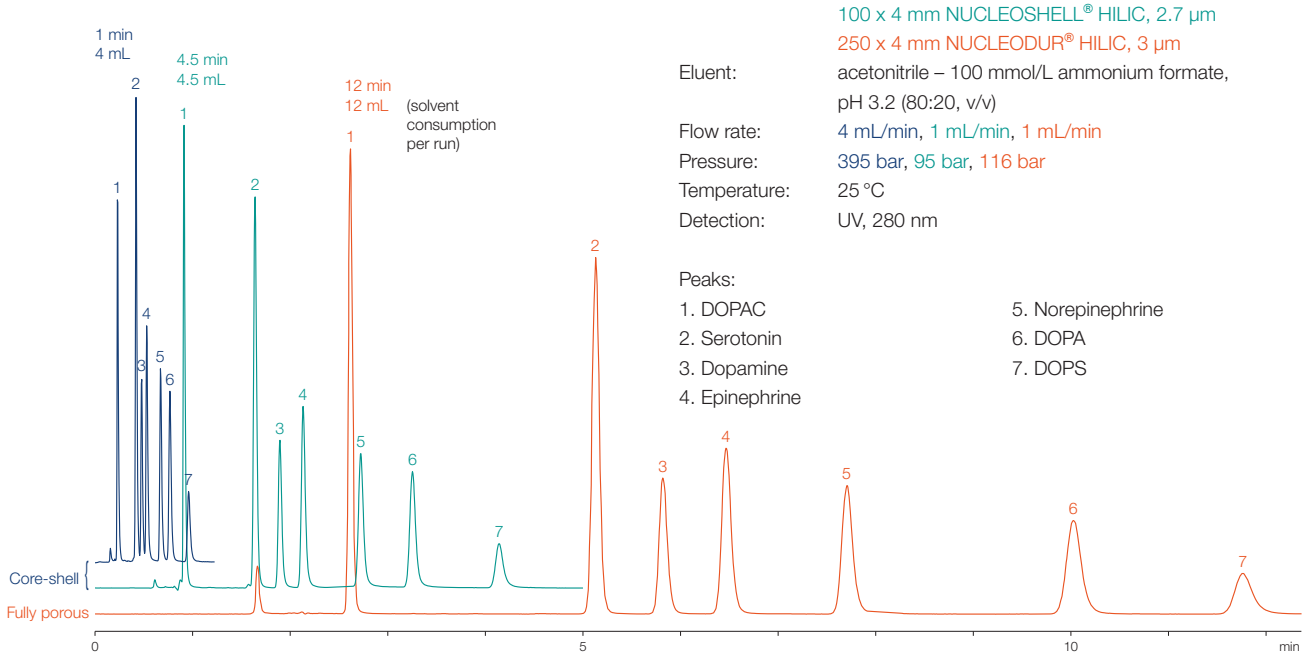


The following chromatograms show the method transfer from a fully porous 3 µm HILIC phase to 2.7 µm core-shell silica with equal selectivity features.

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 %.

Separation of catecholamines

MN Appl. No. 125440



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

Core-shell silica: separation in 1 min pressure < 400 bar

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

Eluent in column acetonitrile – water

ID		Length → 50 mm	100 mm	150 mm	EC guard columns*
NUCLEOSHELL® HILIC, 2.7 µm; particle size 2.7 µm					
Analytical EC columns					
	2 mm	763332.20	763334.20	763336.20	763338.20
	3 mm	763332.30	763334.30	763336.30	763338.30
	4 mm	763332.40	763334.40	763336.40	763338.30
	4.6 mm	763332.46	763334.46	763336.46	763338.30
EC columns in packs of 1, guard columns in packs of 3.					

Guard column system

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

For details of the EC column system please see page 258.