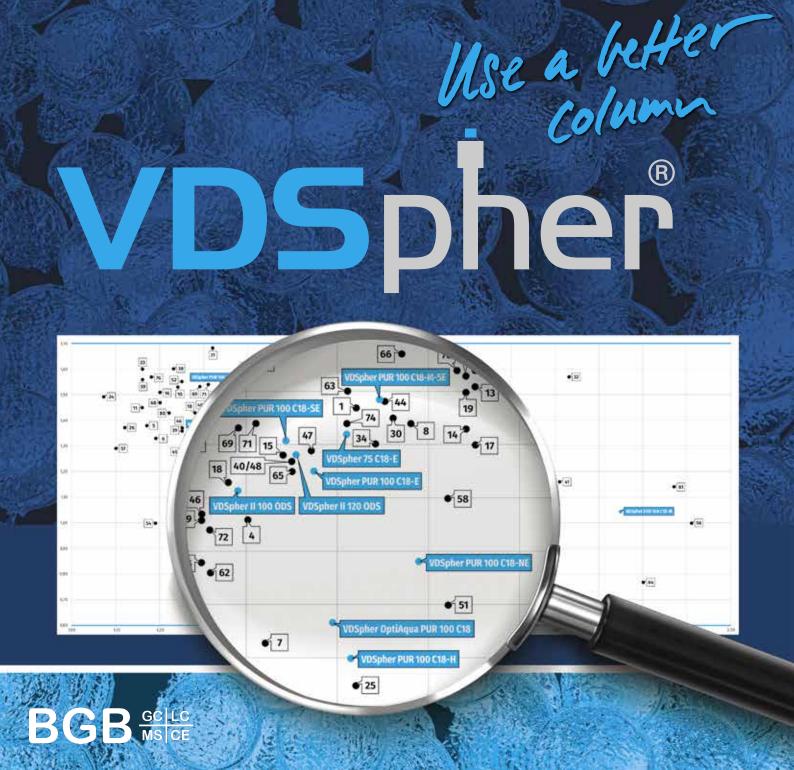
# HIGH PERFORMANCE SEPARATION PHASES HPLC · UHPLC · LC/MS





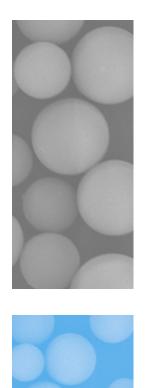
### VDSpher® Chromatography columns "Made in Berlin"

VDS optilab has been producing HPLC columns in all standard and special dimensions, with a variety of separation phases, since 1987. The high quality of our products convinces users all over the world, from universities and regulatory authorities to small and medium-sized companies and large corporations in the chemical and pharmaceutical industries. Our VDSpher separation phases were introduced in 2007. Since then, we have continuously developed the VDSpher product line and can offer a wide range of pore sizes and particle diameters as well as modifications that cover almost all areas of liquid chromatography. Since 2024, we have been offering another new product line, VDSpher II separation phases.

### The extremely extensive VDSpher product range offers solutions for many separation problems.

### **Overview of the various VDSpher separation phase groups:**

4	Page	VDSpher <sup>®</sup> Classic and PUR
6	Page	VDSpher <sup>®</sup> Normal phases
7	Page	VDSpher <sup>®</sup> Reversed phases
13	Page	U-VDSpher® PUR
14	Page	Selectivity map
16	Page	VDSpher <sup>®</sup> II
19	Page	VDSpher <sup>®</sup> CSM
20	Page	VDSpher® OptiAqua
22	Page	VDSpher <sup>®</sup> OptiBio
24	Page	VDSpher <sup>®</sup> PUR HILIC
26	Page	VDSpher <sup>®</sup> Preparative





The standard particle diameters available range from 1.8  $\mu m$  to 10  $\mu m$ :

- Classic: 5 μm and 10 μm
- PUR: 1.8 μm, 2.5 μm, 3.0 μm, 3.5 μm, 4.0 μm, 5.0 μm and 10 μm,
- larger particles are available on request,
- selected modifications are offered in a variety of particle sizes.

The same selectivity from analytical to preparative chromatography (from UHPLC to preparative HPLC) enables targeted upscaling from analytical HPLC via semi-preparative/preparative HPLC to separation on a production scale. Downscaling from analytical HPLC to UHPLC is also simple and straightforward.

VDSpher separation phases with a pore size of 100 or 120 Å are suitable for analysing analytes with small to medium molecular weights. Silica gel phases with a pore size of 300 Å are available for analysing larger molecules.

### Benefit from the advantages of our VDSpher phases:

- very good reproducibility from batch to batch
- broad selectivity spectrum
- very high purity
- narrow pore size distribution
- narrow particle size distribution



### Silica gel base: VDSpher® Classic and VDSpher® PUR

VDSpher Classic and VDSpher PUR phases are based on two different base silica gels, which differ essentially in their purity. The physical specifications of the two base silica gels are summarised in **Table 1**.

The purity of the base silica gel has a noticeable influence on chromatographic separation, as the metal content changes the properties of the silica gel and the surface (see **Figure 1**). Therefore, the interactions between the metal ions and the electron-rich analytes cannot be neglected, as shown in **Figure 2**  are shown. Due to the higher metal content of the VDSpher Classic 100 C18-E phase, the analytes were more strongly retarded compared to VDSpher PUR 100 C18-E.

All VDSpher Classic and PUR silica gels are suitable for analytical, semi-preparative and preparative scale. To achieve the best possible column packing, we recommend VDSpher PUR phases for analytical applications and VDSpher Classic phases for preparative applications.

### Table 1: Physical properties of VDSpher® Classic and PUR

	VDSpher Classic		VDSph	er PUR
Pore size	100	100 300 100		300
Surface [m²/g]	320	90	320	90
Pore volume [mL/g]	0.8	0.8	0.8	0.8
Si concentration [%]	99.97	99.97	99.995	99.995
Metal content [ppm]	< 100	< 100	< 20	< 20
Density [g/mL]	0.45	0.45	0.45	0.45

Other pore sizes on request

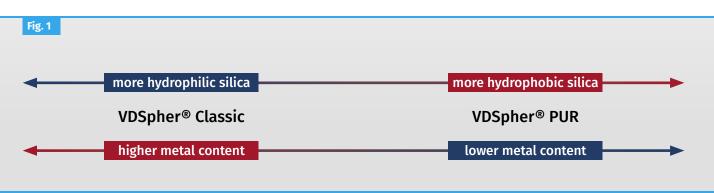


Fig. 1: Influence of the metal content on the hydrophobicity of the base silica gel



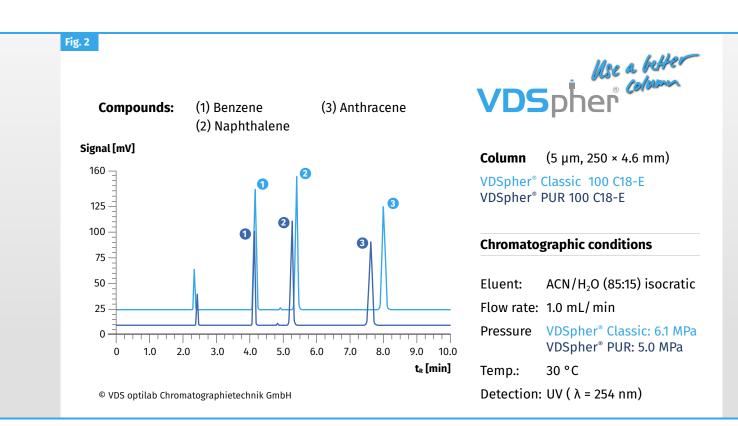


Fig. 2: Influence of the purity of the base silica gel on retention: comparison of VDSpher® 100 C18-E and VDSpher® PUR 100 C18-E

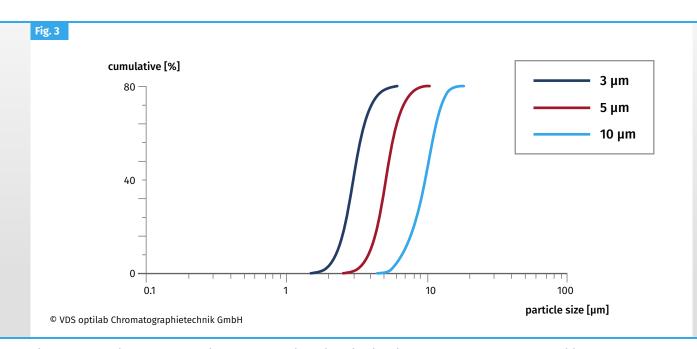


Fig. 3: Cumulative representation of the particle size distribution of three VDSpher® PUR silica gels

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### VDSpher® Normal phases

Both unmodified and modified silica gels are available for normal phase chromatography (NP), enabling a wide range of normal phase separations. **Table 2** provides an overview of the specifications of the available VDSpher Classic and PUR normal phase modifications.

### Table 2: VDSpher® Classic & PUR normal phase modifications and physical specifications

Phase	Modification	Endcapping	Carbon content [%]	pH range	USP Code
SIL	None	-	-	2 – 8	L3
Diol	Dihydroxyalkyl	no	4.5	2 – 8	L20
CN	Alkylnitrile	liquid	6.5	2 – 8	L10
CN-SE (PUR)*	Alkylnitrile	gas	7.0	2 – 9	L10
NH2	Alkylamine	no	4.0	2 - 7.5	L8
NH2-SE (PUR)*	Alkylamine	gas	5.0	2 - 7.5	L8

\*(PUR) only available on silica gel base VDSpher PUR.

The silanol groups on the surface of the silica gels have a slightly acidic reaction, while amino modifications have an alkaline effect. In contrast, diol and CN are neutral modifications.

### Depending on the type of modification, different mechanisms can be used for separation:

#### SIL:

polar interactions, high silanophilic activity

#### Diol:

polar interactions, hydrogen bonds, silanophilic activity

### CN:

polar interactions,  $\pi\text{-}\pi$  interactions, hydrophobic interactions, very low silanophilic activity

#### CN-SE:

polar interactions,  $\pi$ - $\pi$  interactions, hydrophobic interactions, no silanophilic activity

#### **NH2:**

polar interactions, hydrophobic interactions, ion exchange, silanophilic activity

#### NH2-SE:

polar interactions, hydrophobic interactions, ion exchange, no silanophilic activity



### VDSpher® RP phases

Reversed phase chromatography (RP) is the most commonly used HPLC method. As a result, the number of available reversed phases is constantly increasing so that a suitable stationary phase is available for every separation problem. This means, for example, that C18 is no longer just C18: many different modification and endcapping approaches lead to a wide range of very different C18 phases.

A general overview can be found in **Table 3**. In addition, there are further modifications in our VDSpher product line with VDSpher OptiAqua and OptiBio (p. 20 and 22) as well as the VDSpher II ODS phases (p. 16).

### Table 3: VDSpher® Classic and PUR RP phase modifications and physical specifications

Phase	Modification	Carbon content [%]	Endcapping	pH range	USP Code
C18-NE (PUR)*	C18	16.3	no	2 - 7.5	L1
C18-E	C18	16.8	liquid	2 - 7.5	L1
C18-SE	C18	17.0	gas	2 – 9	L1
C18-M	C18	17.5	no	2 – 8	L1
С18-М-Е	C18	19.0	liquid	2 – 8	L1
C18-M-SE	C18	20.0	gas	1 – 9	L1
C18-H	C18	11.5	polar	2 - 7.5	L1
C8-NE (PUR)*	C8	9.9	no	2 - 7.5	L7
С8-Е	C8	10.0	liquid	2 - 7.5	L7
C8-SE	C8	10.4	gas	2 – 9	L7
C8-M (PUR)*	C8	10.0	no	2 – 8	L7
C8-M-E (PUR)*	C8	10.7	liquid	2 – 8	L7
C8-M-SE (PUR)*	C8	11.0	gas	2 – 9	L7
C8-H (PUR)*	C8	8.5	polar	2 - 7.5	L7
C4-E	C4	7.0	liquid	2 - 7.5	L26
Phenyl-E	Alkylphenyl	10.5	liquid	2 - 7.5	L11
Phenyl-SE (PUR)*	Alkylphenyl	10.7	gas	2 – 9	L11
Phenyl-Hexyl (PUR)*	Alkylphenyl	14.0	liquid	2 - 7.5	L11
Bi-Phenyl (PUR)*	Alkylphenyl	12.8	liquid	2 - 7.5	L11

\*(PUR) only available on silica gel base VDSpher PUR.

Depending on the type of reagents used, a single ('monomeric') or multiple ('polymeric') bonding to the surface silanol groups of the base silica gel is achieved in the modification step, which then leads to a brush-like or branched structure. For steric reasons, however, not all silanol groups can be modified in both cases, so that the phases still show a noticeable silanophilic activity. To reduce or completely avoid this, endcapping with trimethylchlorosilane is carried out in a subsequent step. The reaction control of the endcapping also plays a role here: in the liquid phase, only approx. 40 % of the remaining silanols can react with trimethylchlorosilane; in the gas phase, this figure is up to 99 %. In another variant, a reagent with a polar group is used in addition to the monomeric modification, resulting in a mixed phase. The special properties of this phase are described in detail below.

### The different VDSpher C18 phases result from the combination of the modification and endcapping methods described above:

C18-NE	monomeric bonding	no endcapping
C18-E	monomeric bonding	Liquid phase endcapping
C18-SE	monomeric bonding	Gas phase endcapping
C18-M	polymeric bonding	no endcapping
С18-М-Е	polymeric bonding	Liquid phase endcapping
C18-M-SE	polymeric bonding	Gas phase endcapping
С18-Н	monomeric bonding	Mixed phase

In addition, the polar group in the VDSpher Classic 100 C18-H phase can be used to achieve a different selectivity: **Figure 4** shows the purification of insulin (peak 2) with the phases VDSpher Classic 100 C8-E and VDSpher Classic 100 C18-H. For this example, the phase VDSpher Classic 100 C8-E was chosen as it has a comparable carbon load to the phase VDSpher Classic 100 C18-H. It should be noted that when using VDSpher Classic 100 C8-E, the insulin elutes between its impurities, whereas in the 'polar' phase VDSpher Classic 100 C18-H the retardation of the insulin is longer than for the impurities.



Purification of Insulin Using Different Reversed Phase Modifications with Comparable Carbon Load

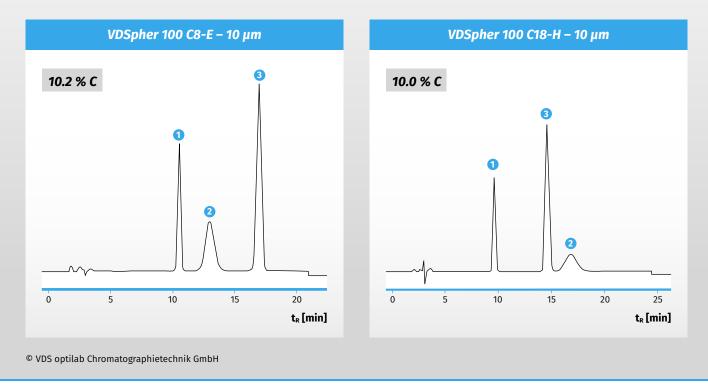


Fig. 4: Purification of insulin with different VDSpher® Classic 100-RP phases

Normally, the standard phase VDSpher C18-E is a very good starting point for many separation problems. It is of medium hydrophobicity and shows low silanophilic activity. VDSpher C18-E has therefore proven itself in many applications, e.g. in the isolation of natural substances, in the determination of caffeine in coffee, tea and other caffeinated beverages and even in the determination of amino acids. The VDSpher C18-SE phase, which shows no silanophilic activity, is slightly more hydrophobic. Hydrophobic substances are therefore more strongly retarded by this phase. In addition, the complete endcapping and the resulting inertness make it easier to analyse alkaline substances and achieve increased stability in strongly acidic and slightly alkaline media. On the other hand, the VDSpher C18-NE phase is available without endcapping if silanophilic activity is important for the separation. The free silanol groups also increase the water mobility compared to the endcapped phases.

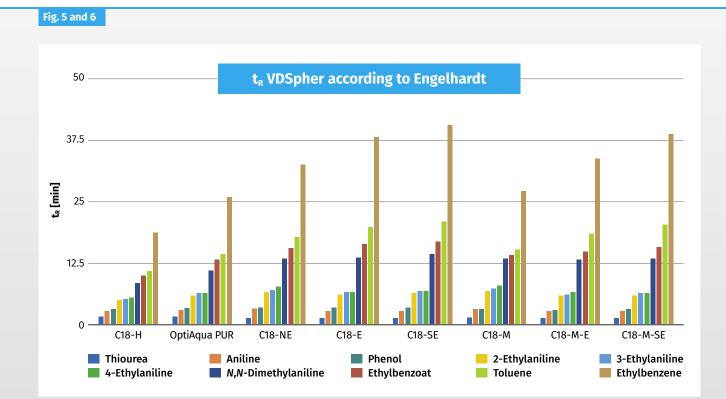
For work in 100 % water as a mobile phase, we recommend the very hydrophilic phase VDSpher C18-H. As this is a mixed phase, the C18 chains do not collapse despite the high water content. VDSpher C18-H is therefore ideal for analysing polar analytes and smaller water-soluble biomolecules.

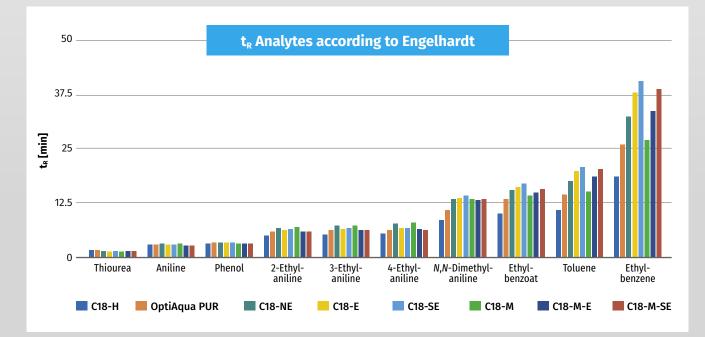
The three phases VDSpher C18-M, VDSpher C18-M-E and VDSpher C18-M-SE can be selected for very hydrophobic applications. These phases have a very high carbon loading. The branched surface structure effectively shields the surface of the silica gel, so that it is possible to work with 100 % water as eluent despite the high hydrophobicity. As with the single bonded phases, VDSpher C18-M (non-endcapped), VDSpher C18-M-E (liquid phase endcapping) and VDSpher C18-M-SE (gas phase endcapping) are available in three modifications so that the influence of silanol groups and carbon content can be taken into account for the desired application.

The described modifications and their effects are not limited to C18: a large number of different VDSpher phases are also available for C8 and C4 modifications. Due to the lower carbon loading compared to C18 phases, C8 and especially C4 phases are less hydrophobic. In addition, the silanophilic activity is more pronounced due to the better accessibility of the silica gel surface.

VDSpher Phenyl-E, Phenyl-Hexyl and Bi-Phenyl with liquid phase endcapping and VDSpher Phenyl-SE with gas phase endcapping are alternatives to the aliphatic modified reversed phases.  $\pi\pi$ -interactions influence the separation through the alkylphenyl modification and enable other selectivities, e.g. for polar and non-polar aromatic hydrocarbons or fatty acids.

To visualise the selectivities, the Engelhardt test was carried out with various VDSpher phases, as shown in **Figure 5**. This test named after Engelhardt\* with eight compounds is one of the standard tests for assessing the hydrophobicity and silanophilic activity of reversal phases. While toluene and ethyl benzene exhibit hydrophobic properties, phenol and ethyl benzoate produce neutral polar interactions. The critical behaviour towards basic compounds is assessed by injecting five different anilines. If the silanol activity of the stationary phase is well suppressed, aniline elutes before phenol, and the geometric isomers of ethyl aniline coelute. \* H. Engelhardt, M. Jungheim, *Chromatographia* **29**, 59 – 68 (1990)





MeOH/H $_2$ O (55:45), 5  $\mu$ m, 150 × 4.6 mm, flow 1.0 mL/min, UV 254 nm, ambient temperature

Fig. 5 and 6: Engelhardt test on different VDSpher phases

### U-VDSpher® PUR phases



More and more users are utilising UHPLC (Ultra High Performance Liquid Chromatography) as it offers short analysis times, high efficiency and therefore a high level of cost-effectiveness.

Suitable packing materials and columns must fulfill certain requirements for UHPLC. Small column dimensions lead to short analysis times, but the desired high chromatographic resolution cannot be achieved with the particle sizes commonly used in HPLC. In order to achieve the desired resolution, silica gels with particle diameters of less than 2  $\mu$ m ('sub-2- $\mu$ m particles') were developed. This results in very high plate numbers, which leads to a very high resolution.

The available modifications based on U-VDSpher PUR 100 are shown in **Table 4**.

### Table 4: Physical specifications and modifications of U-VDSpher® PUR 100, 1.8 µm

Phase	Modification	Endcapping	Carbon content [%]	USP Code
SIL	none	-	-	L3
C8-E	C8	liquid	10.0	L7
C18-E	C18	liquid	16.8	L1
C18-SE	C18	gas	17.0	L1
C18-M	C18	no	17.5	L1
C18-M-SE	C18	gas	20.0	L1
С18-Н	C18	polar	11.5	L1
CN	Alkylnitrile	liquid	6.5	L10
Phenyl-E	Alkylphenyl	liquid	10.5	L11







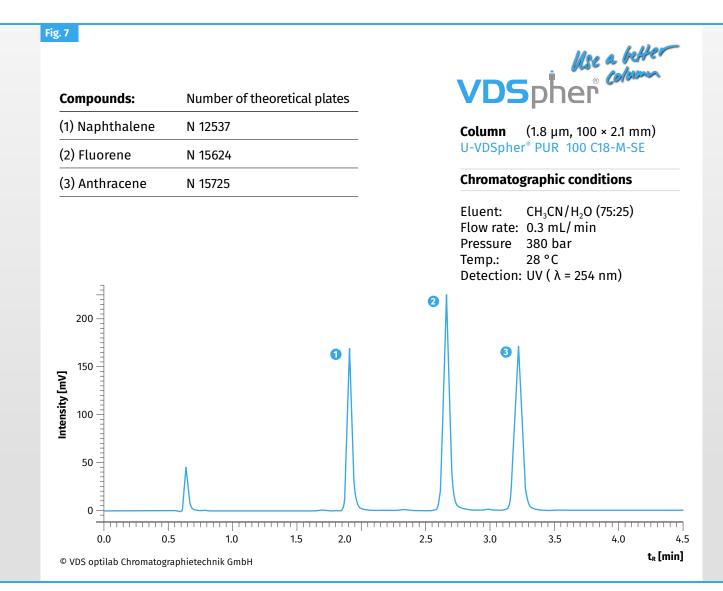
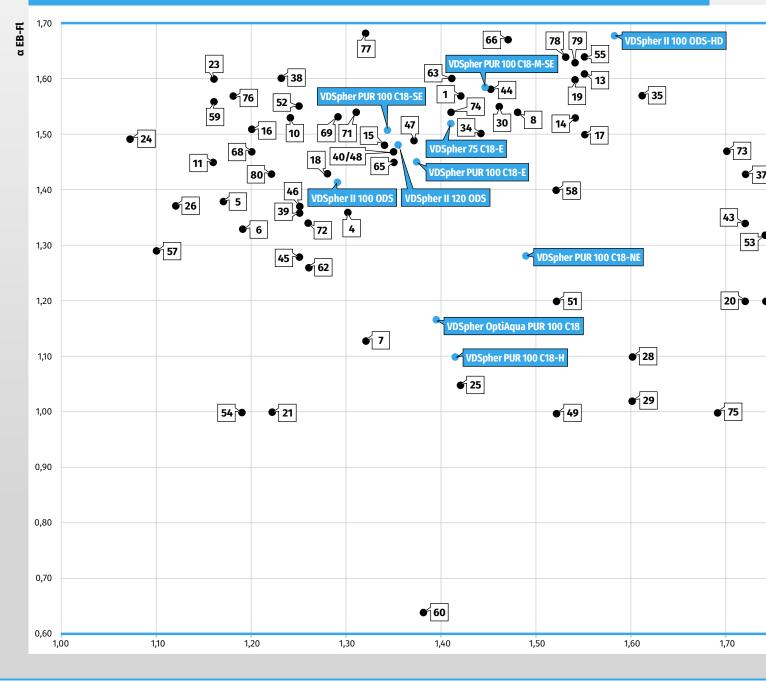


Fig. 7: Test chromatogram U-VDSpher PUR 100 C18-E



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### Selectivity card α EB-Fl/Tri-oTer

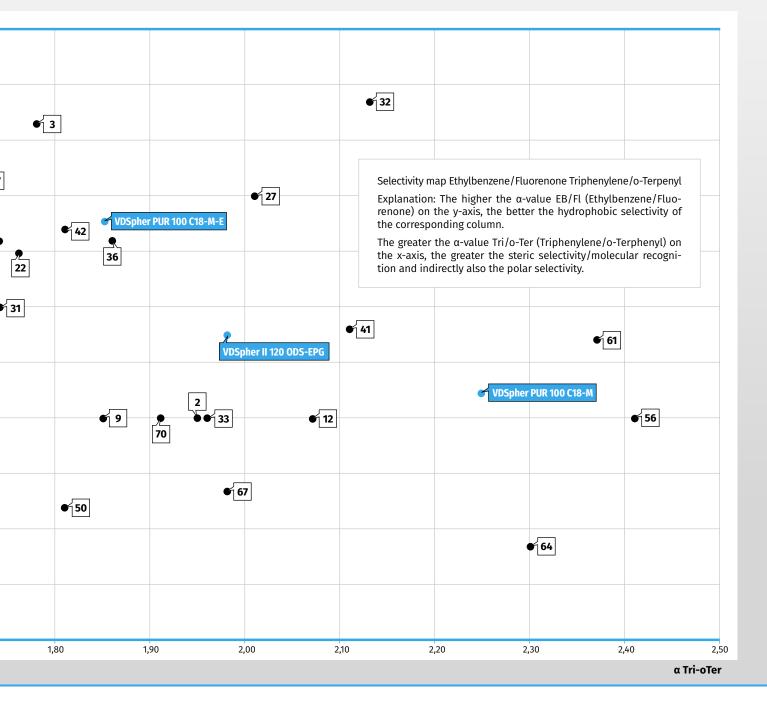


Column	No
PerfectSil Target ODS-3HD	35
Polaris C18-A	36
Polaris C18-Ether	37
Prodigy ODS 2	38
ProntoSil 120-5-AQ	39
ProntoSil 120-5-C18-H	40
ProntoSil 120-5-ace-EPS	41
Purospher RP-C18 endcapped	42
Purospher STAR RP-18 endcapped	43
Pursuit C18	44
Raptor ARC-18	45
ReproSil-PUR AQ	46
ReproSil-PUR ODS 3	47
Reprospher C18 DE	48
Reprospher C18-Aqua	49
Spherisorb ODS 1	50
Spherisorb ODS 2	51

Column	No
Inertsil ODS 3	18
Kromasil C18	19
LiChrosorb C18	20
LiChrospher 60 RP-Select B	21
LiChrospher RP-18	22
Luna C18 (2)	23
Luna Omega C18	24
NUCLEODUR C18 Gravity	25
NUCLEODUR C18 Gravity SB	26
NUCLEODUR C18 Isis	27
NUCLEODUR Sphinx RP	28
NUCLEOSIL 100-5 C18	29
NUCLEOSIL 100-5 C18 HD	30
NUCLEOSIL 50-5 C18 ec	31
NUCLEOSIL C18 AB	32
NUCLEOSIL C18 Nautilus	33
Nova-Pak C18	34

Column	No
ACQUITY UPLC BEH C18	1
Acclaim 120 C18	2
Alltima HP C18	3
Aqua C18	4
Atlantis T3	5
Atlantis dC18	6
µBondapak C18	7
Discovery C18	8
Discovery RP Amid C16	9
Gemini C18	10
Gemini NX-C18	11
HyPURITY Advance	12
HyPURITY C18 HCT	13
Hypersil BDS	14
Hypersil GOLD	15
InertSustain C18	16
Inertsil ODS 2	17

Selectivity map



Column	No
SunFire C18	52
Superspher 100 RP C18	53
Superspher 60 RP-Select B	54
Symmetry C18	55
Symmetry Shield RP C18	56
Synergi Fusion-RP	57
Synergi Hydro-RP	58
Synergi MAX RP	59
Synergi POLAR RP	60
Ultrasep ES RP 18E	61
VYDAC C18 201HS	62
XBridge C18	63
XBridge Shield RP C18	64
XSelect CSH C18	65
XSelect HSS C18	66
XSelect HSS C18 SB	67
XSelect HSS T3	68

Column	No
XTerra MS C18	69
XTerra RP C18	70
YMC Pro C18	71
YMC-Pack ODS-AQ	72
YMC-Triart C18 ExRS	73
YMC-Ultra HT Pro C18	74
ZORBAX Bonus RP	75
ZORBAX Eclipse Plus C18	76
ZORBAX Eclipse XDB C18	77
ZORBAX Extend C18	78
Zorbax ODS	79
Zorbax SB-C18	80

Our special thanks go to Dr Stavros Kromidas, Blieskastel, for providing the values for the creation of this selectivity map. Some separation phases have been omitted for reasons of clarity. The complete overview, as well as other selectivity maps and more: www.kromidas.de or www.colona.kromidas.de

## VDSpher® II phases

The new VDSpher II separation phases are both an independent silica line and an excellent addition to VDSpher PUR and Classic. This opens up additional options for better substitution of foreign phases.

VDSpher II is also based on high-purity silica gel (99.999 %). The 100 Å variant has a large chromatographic surface area and a large pore volume. It is suitable for smaller molecules, but also as a standard ODS phase. VDSpher II is available in two pore sizes and two particle sizes.

### Table 5: Physical specifications of VDSpher® II reversed phases

	VDSpher 100	VDSpher 120
Pore size [Å]	100	120
Surface [m²/g]	450	300
Pore volumes [mL/g]	1.1	1.0
Si concentration [%]	99.999	99.999
Metal content [ppm]	< 10	< 10





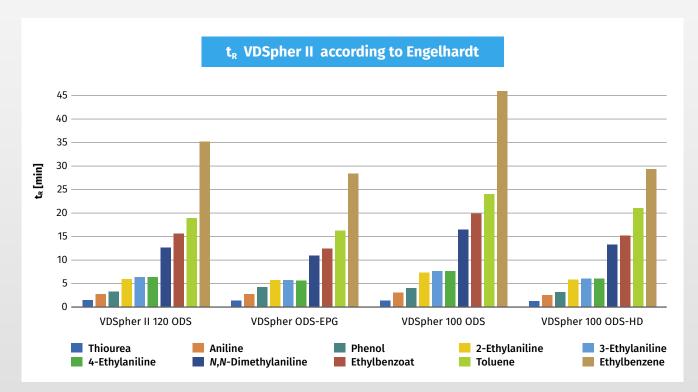
VDSpher II has excellent chemical and mechanical stability. The carefully controlled complete endcapping shows best separation properties for acidic, basic and chelating compounds, making it the ideal choice for a wide range of organic compounds. The ODS modifications are excellent all-rounders for an extremely wide range of HPLC applications. Due to its high carbon content and excellent endcapping, the ODS-HD modification is very well suited for extreme pH values and shows brilliant peak sharpness. ODS-EPG (Embedded Polar Group) with the polar embedded group has a different selectivity than normal ODS phases and is therefore also suitable for 100 % aqueous applications and polar analytes.

### Table 6: VDSpher® II phases

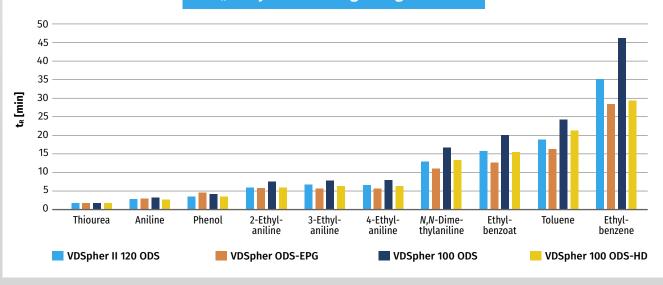
Phase	Particle size [µm]	functional	Endcapping	Carbon content [%]	pH range	USP Code
100 ODS	3 & 5	mono	fully	17.0	1.5 – 9	L1
100 ODS-HD	3 & 5	mono	fully	24.0	1 – 10	L1
120 ODS	3 & 5	mono	fully	17.0	2 – 8	L1
120 ODS-EPG	3 & 5	mono	fully	18.5	2 - 7.5	L1 / L60



Fig. 8



t<sub>R</sub> analytes according to Engelhardt



MeOH/H<sub>2</sub>O (55:45), 5 µm, 150 × 4.6 mm, flow 1.0 mL/min, UV 254 nm, ambient temperature, s. page 10/11





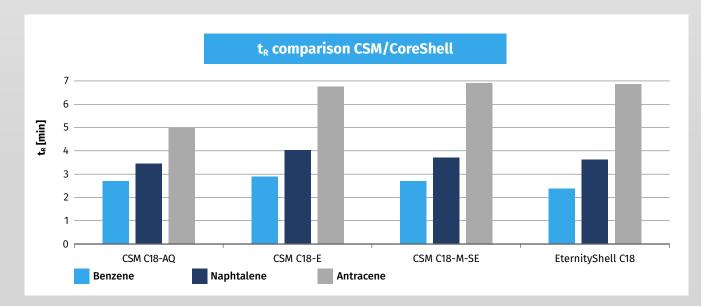
### VDSpher<sup>®</sup> CSM (Core Shell Mode) phases

The principle of core shell phases is another very interesting development in HPLC. With a non-porous core and a thin chromatographic layer, plate numbers of materials smaller than 2  $\mu$ m can be achieved from materials with a particle diameter of 2.5 – 2.7  $\mu$ m. The reason for this is the faster diffusion of the analytes. The resulting back pressure is comparable to the separation columns packed with 3  $\mu$ m material. The disadvantage is the lower chromatographic area in a small space and the loadability of the thin chromatographic layer. The VDSpher Core Shell Mode Material (CSM) takes a different approach to achieve similar results. It is a fully porous 2.5  $\mu$ m silica gel with an optimised pore diameter. This results in a smaller chromatographic surface area, which enables faster diffusion than 100 Å particles. A pleasant side effect is the higher loadability of the Core Shell Mode Phases.

### Table 7: VDSpher® CSM phases

Fig. 9

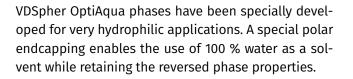
Phase	Carbon content [%]	pH range
C18-E	6.5	2 - 7.5
C18 M-SE	10	2 - 9
C18 AQ	6	2 - 7.5



ACN/H<sub>2</sub>O (65:35), 5  $\mu$ m, 150 × 4.6 mm, flow 1.0 mL/Min, ambient temperature

Fig. 9 : Comparison of VDSpher® CSM phases vs. core shell

### VDSpher® OptiAqua phases



Based on VDSpher Classic and PUR, four VDSpher OptiAqua phases with different modifications and particle diameters have been developed, which are suitable for separations of polar and non-polar analytes. This provides ideal conditions for both preparative and analytical HPLC.

The available VDSpher OptiAqua and OptiAqua PUR reversed phase modifications are shown in **Table 8**.

All VDSpher OptiAqua phases are pH-stable in a range from 2 to 7.5 and can be used up to a temperature of 60 °C without any problems.

### Table 8: VDSpher® OptiAqua and OptiAqua PUR modifications

Phase	Pore size [Å]	Carbon content [%]	pH value [g/mol]	USP Code
OptiAqua 100 C18	100	13.0	30 - 800	L1
OptiAqua 100 C8	100	8.1	30 - 800	L7
OptiAqua PUR 100 C18	100	13.0	30 - 800	L1
OptiAqua PUR 100 C8	100	8.1	30 - 800	L7

### **Possible applications are e.g:**

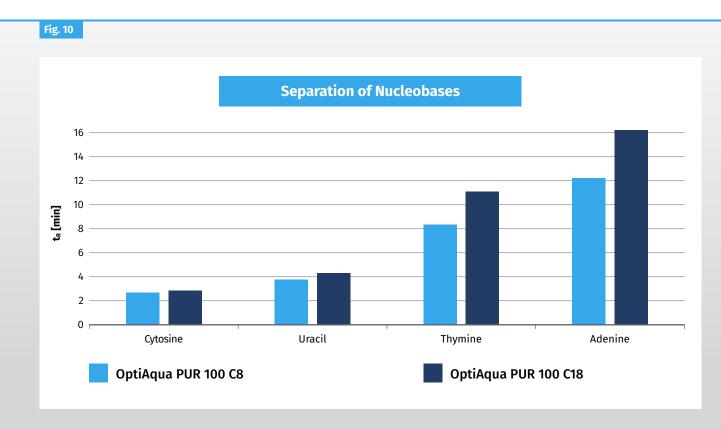
- Antibiotics
- Biomolecules
- Drugs
- Nucleobases
- organic acids

- Parabens
- Pesticides
- Sulfonamides
- water-soluble vitamins
- Xanthine









 $KH_2PO_4, 5~\mu m,\,150~\times$  4.6 mm, flow 1.0 mL/min, ambient temperature

Fig. 10: Separation of nucleobases with VDSpher OptiAqua PUR

### VDSpher® OptiBio phases

VDSpher OptiBio phases have been specially developed for bio-chemical analyses, e.g. protein and peptide separation. Various separation phases with different modifications are available in 5  $\mu$ m, which

are suitable for a wide range of applications. (10  $\mu$ m on request). **Table 10** provides an overview of the VDSpher OptiBio PUR reversed-phase modifications and their physical specifications.

### Analytes that can be analysed with VDSpher OptiBio columns are, for example

Antibodies

- Oligonucleotides
- Peptides

- Proteins
- other biomolecules





### Table 9: Pore size according to the molar mass range of the analytes

Pore size [Å]	Spherical analytes [g/mol]	Cylindrical analytes [g/mol]
300	800 - 21500	350 – 9500

#### Table 10: VDSpher® OptiBio PUR/Classic phase modifications

Phase	Pore size [Å]	Endcapping	pH range	Max. H2O proportion [%]	USP Code
OptiBio C18-M	300	no	2 - 8	100	L1
OptiBio PUR C18-E	300	liquid	2 - 7.5	95	L1
OptiBio PUR C18-SE	300	gas	2 – 9	95	L1
OptiBio PUR C18-M-E	300	liquid	2 - 8	100	L1
OptiBio PUR C18-M-SE	300	gas	1 – 9	100	L1
OptiBio PUR C8-E	300	liquid	2 - 7.5	95	L7
OptiBio PUR C8-SE	300	gas	2 – 9	95	L7
OptiBio PUR C8-M-E	300	liquid	2 - 8	100	L7
OptiBio PUR C8-M-SE	300	gas	2 – 9	100	L7
OptiBio PUR C4-E	300	liquid	2 - 7.5	95	L26
OptiBio PUR C4-SE	300	gas	2 – 9	95	L26
OptiBio PUR C4-M-E	300	liquid	2 - 8	100	L26
OptiBio PUR DIOL	300	no	2 - 8	100	L20

All VDSpher OptiBio modifications are stable over a pH range of pH 2 – 7.5 and can be used up to a temperature of 60 °C without any problems. Eluents with a high water content are suitable for all VDSpher OptiBio phases. The modifications and C18-M-SE and C18-SE are very hydrophobic, C18-M-E, C18-E and C18-M have a medium hydrophobicity.

### VDSpher<sup>®</sup> PUR HILIC phases



HILIC (Hydrophilic Interaction Liquid Chromatography) is a special variant of HPLC in which hydrophilic normal phase modifications are used with mobile phases of reversed phase chromatography. This promotes the separation of very polar substances that are otherwise very difficult to analyse using proven HPLC methods.

In HILIC mode, an aqueous layer is formed on the surface of the stationary phase (see **Fig. 11**). The separation of the analytes is based on a complex combination of different effects, as in addition to the usual interactions, the distribution of the analytes between the mobile phase and the water layer also has a crucial influence.

The available VDSpher PUR HILIC phases are listed in **Table 11**. The various available modifications allow the optimum phase to be selected for each HILIC application. For example, VDSpher PUR HILIC-AM and especially VDSpher PUR HILIC-SAC are ideal for sugar separations, while VDSpher PUR HILIC-Z is ideal for analysing nucleobases.

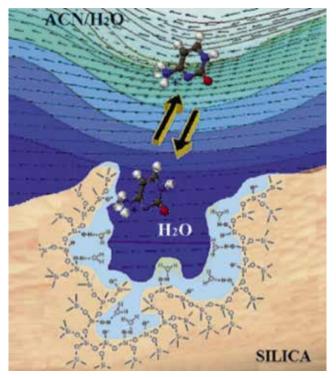


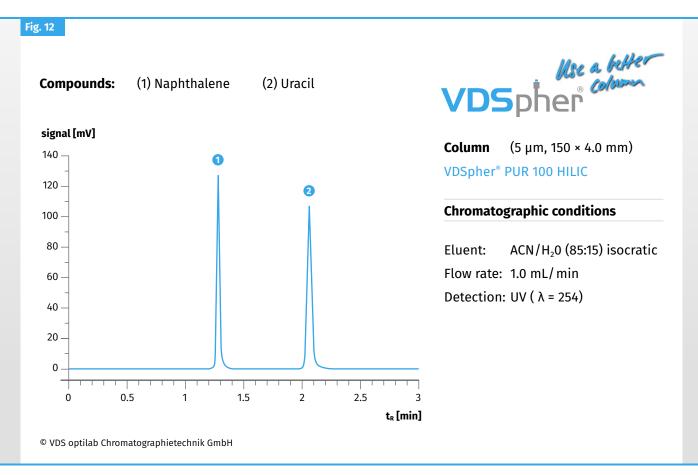
Fig. 11: The figure was kindly provided by Bogusław Buszewski (Environmental Chemistry and Bioanalytics, Nicolaus Copernicus University, Toruf, Poland) B. Buszewski, S. Noga, Anal Bioanal Chem. 402, 231 – 247 (2012)

Phase	Particle size [µm]	Pore size in Å	Modification	USP Code
HILIC	5	100	None	L3
HILIC-OH	5	100	Dihydroxyalkyl	L20
HILIC-AM	5	100	Alkylamine	L8
HILIC-SAC	5	100	Alkylamine	L8
HILIC-Z	5	100	Zwitterionic	_

#### Table 11: VDSpher® PUR 100 HILIC phases



The ability to retard polar substances is one of the particular strengths of HILIC. For example, Uracil, which is often used as a dead time marker in the HPLC is strongly retarded in HILIC mode, as shown in **Figure 12**.





### VDSpher<sup>®</sup> for preparative applications

VDSpher has proven itself not only on an analytical scale, but also for semi-preparative and preparative HPLC. The excellent scalability allows easy switching from small particle diameters (1.8  $\mu$ m to 5  $\mu$ m) to larger particle sizes (10  $\mu$ m, > 10  $\mu$ m on request) which are favoured in semi-preparative and preparative applications.

All VDSpher Classic and PUR silica gels can be used on a semi-preparative and preparative scale. In order to achieve the best possible column packing, we recommend VDSpher<sup>®</sup> Classic separation phases especially for columns with an inner diameter of 30 mm and larger. **Table 12** provides information on the dimensions of preparative columns.

### Table 12: Column dimensions for semi-preparative and preparative applications

Inner diameter [mm]	Column lengths [mm]
8.0	100 / 150 / 250
10.0	150 / 250
20.0	30 / 150 / 250
30.0	100 / 150 / 250
50.0	100 / 150 / 250

Other column lengths and inner diameters as well as refill (cost-saving, environmentally friendly) on request

Some VDSpher PUR modifications are available in a particle size of 7  $\mu$ m. This represents a valuable compromise between 5 and 10  $\mu$ m:

Less pressure than with 5  $\mu m$  – higher plate numbers than with 10  $\mu m$  particle size.

### Table 13: Comparison of back pressure and plate number for VDSpher® PUR 100 C18-E 5, 7 and 10 µm

Particle size [µm]	Pressure [bar]	Number of theoretical plates per meter
5	48	100 000
7*	25	70 000
10	14	45 000

\*On request





VDS optilab Chromatographietechnik GmbH endeavours to fulfill the wishes of its customers. For this reason, we also offer special column dimensions that deviate from the standard. Send us your enquiry and we will be happy to make you an offer.

Benefit from our professional customer and application support. Our experts will answer your enquiries quickly and easily, help you select the ideal VDSpher column for your application and support you with application optimisation and column use.

Take advantage of our special offers: Let us arrange your individual test kit for method development and validation. These kits usually contain five different acids at an attractive price and enable you to screen quickly and easily to select the ideal phase for your application.

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