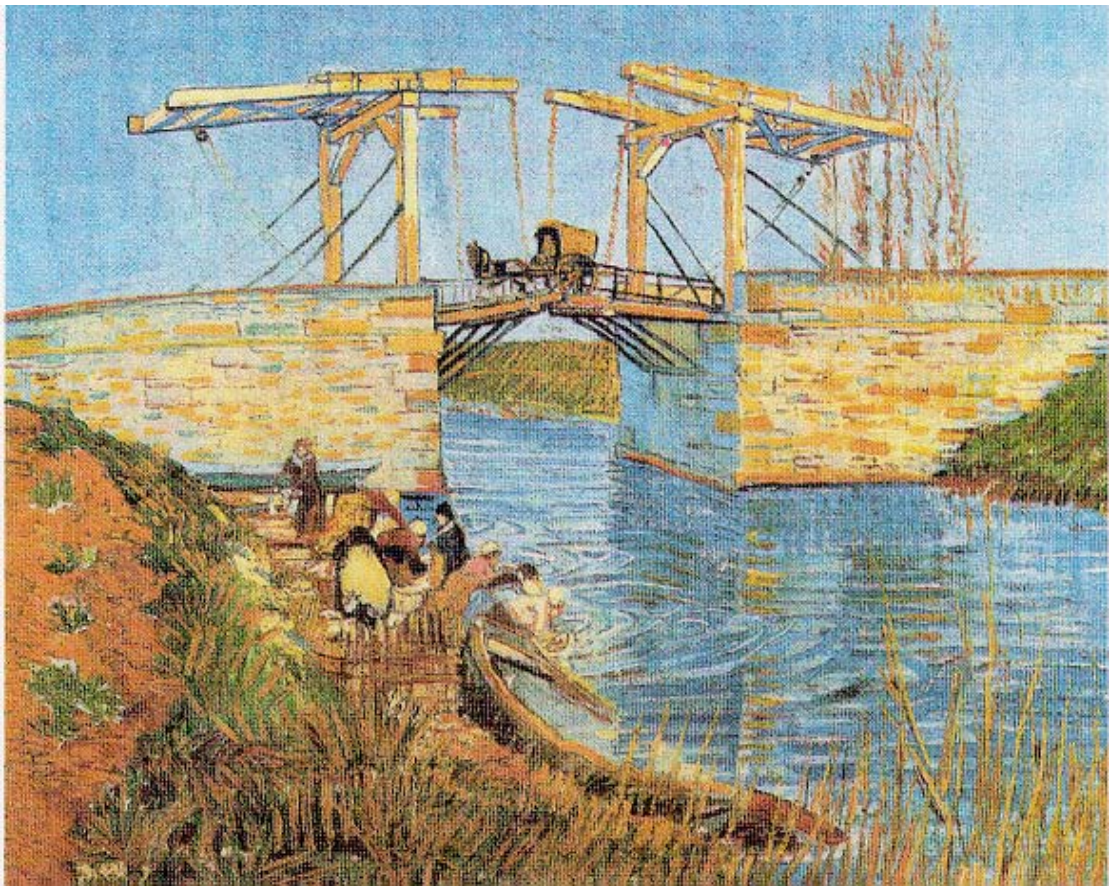


SEPSERV

SEPARATION SERVICE BERLIN
Vertriebsgesellschaft Dr. Eppert & Co.

A bridge to solve your problems

HPLC

English Catalogue 2011

Dear customer,

This catalogue in English includes apart from Part A primarily Part B of our main catalogue. Part B concerns our Special Columns and a lot of examples for application.

We hope, the catalogue will be of benefit, and help you, solving your problems in HPLC.

All columns are packed with UltraSep ES, a spherical silica based on our own development and production with 25 years of experience. We texture and modify this silica according to the respective applications. All packings are very stable against high pressure and remarkably resistant against hydrolysis. This means high lifetimes of the columns on principle. Polar and chromatographically problematic natural products can be eluted from these packings very symmetrically.

CONTENT

Chiral Analysis C

Ion Exchangers I

Other Special Columns O

Literature (Book Offer) *L*

STANDARD COLUMNS

Packings of UltraSep ES

The spherical silicagel UltraSep ES is produced by SEPSERV Separation Service Berlin. The outstanding quality of this powerful HPLC packings has been demonstrated by resolving a lot of separation problems.

Special properties of UltraSep ES packings

- **extreme chemical purity**
- **high inertness**
- **excellent packing properties (long time stability)**
- **excellent pressure stability (up to 1000 bar)**
- **enhanced hydrolytic resistance**
- **remarkable selectivity**

Standard packings

	Polar phases	*Reversed phases*
UltraSep ES	Silica	C1
	NH2	C8
	CN	C18
	OH	Phenyl*

All packings are delivered only in separation columns.
* Standard: Phenylpropylsilica. Phenylhexylsilica see O - 6

SEPARATION COLUMNS and **PRICES** for **Standard Packings (€/piece) *)**

Filled with **4 µm** standard packings listed above **)

Column size (mm)

250 x 4.6	250 x 4	250 x 3	250 x 2	150 x 4.6	150 x 4	150 x 3	150 x 2	100 x 4.6	100 x 4	100 x 3	100 x 2
200 x 4.6	200 x 4	200 x 3	200 x 2	125 x 4.6	125 x 4	125 x 3	125 x 2	60 x 4.6	60 x 4	60 x 3	60 x 2
320.00	285.00	285.00	295.00	240.00	220.00	220.00	230.00	190.00	175.00	205.00	215.00

Filled with **5**, **6**, **7**, **8**, **9** and **10 µm** standard packings

Column size (mm)

250 x 4.6	250 x 4	250 x 3	250 x 2	150 x 4.6	150 x 4	150 x 3	150 x 2	100 x 4.6	100 x 4	100 x 3	100 x 2
200 x 4.6	200 x 4	200 x 3	200 x 2	125 x 4.6	125 x 4	125 x 3	125 x 2	60 x 4.6	60 x 4	60 x 3	60 x 2
230.00	195.00	195.00	260.00	195.00	175.00	175.00	210.00	155.00	140.00	160.00	190.00

Precolumn Cartridges (Prices per pack = 5 pieces)

Size (mm)

10 x 4.6	10 x 4	10 x 3	10 x 2	5 x 4.6	5 x 4	5 x 3	5 x 2	20 x 4.6	20 x 4	20 x 3	20 x 2
185.00	170.00	170.00	170.00	185.00	170.00	170.00	170.00	210,00	190,00	190,00	190,00

***) Prices for the Special Columns (sectons E, F, B, C, I, O) with 4.6 mm internal diameter on request**
****) 3 µm packings on request**

Columns with endcapped packings has to be calculated with an additional charge of 10.00 €.

For analyses with modern HPLC equipments we recommend the use of **Narrowbore Columns** (2 mm internal diameter).

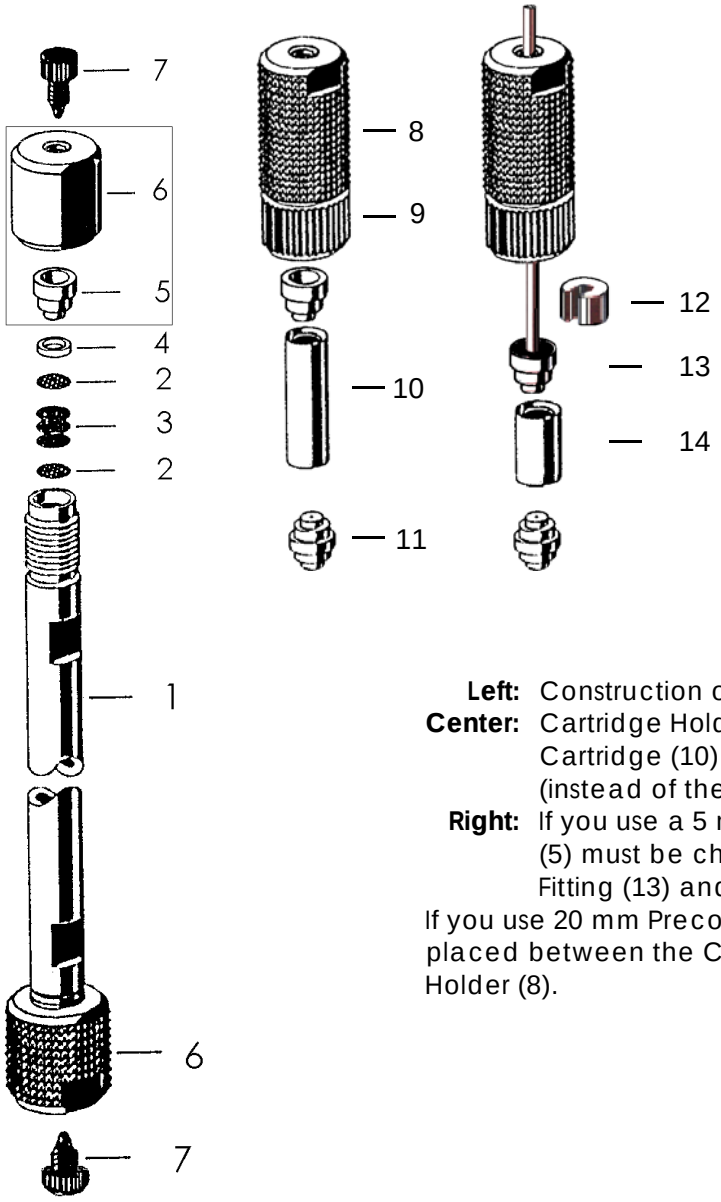
In order to protect the separation columns against contaminations from the samples and the equipment **Precolumn Cartridges** and **Guard Columns** are recommended (see pages 9 and 10).

Columns of 2 – 4 mm internal diameter

SEPSERV Separation Service Berlin offers a **wide range** of HPLC columns.

For 4, 3, and 2 mm diameter SEPSERV uses the HYPERCHROME column system, which is widespread especially in Germany. On request packings will be delivered also in columns of all the other common systems.

Column System HYPERCHROME



1 Column tube
2 Stainless steel sieve
3 Glas fiber filter
4 Teflon ring
5 Column end fitting
6 Column head nut
7 Stopper
8 Cartridge holder
9 Gripp protection
10 Precolumn cartridge 10 mm
11 Double end fitting
12 Distance piece
13 Capillary end fitting
14 Precolumn cartridge 5 mm

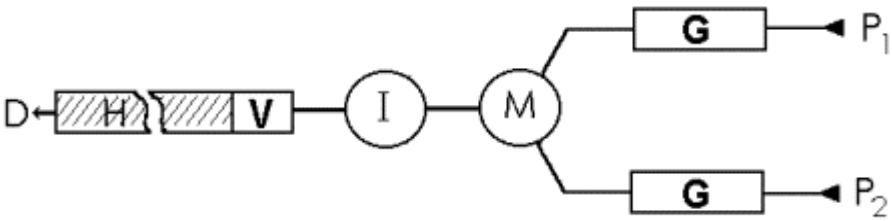
Left: Construction of a HPLC Column
Center: Cartridge Holder (8) with a 10 mm Precolumn Cartridge (10) and a Double End Fitting (11) (instead of the parts (6) and (5) in the left picture)
Right: If you use a 5 mm Precolumn Cartridge (14), part (5) must be changed against the Capillary End Fitting (13) and the Distance Piece (12).

If you use 20 mm Precolumn Cartidges, a lengthening hull is placed between the Column Tube (1) and the Cartridge Holder (8).

Guard Columns
(Protection of HPLC Columns against Mechanical Pollution)

For a trouble-free function of HPLC columns it is essential, to remove solid contaminations from the eluent (buffer solutions!), from samples and equipment (abrasion). This can be achieved by filtration of the eluent and the sample solution and by the usage of **Precolumns** and by **Guard Columns.** Latter are installed into the equipment instead of filters before the injection valve and the mixing chamber, resp.:

G **Guard Columns**
H Separation Column
I Injection Valve
M Mixing Chamber
P Pumps
V **Precolumn**
D Dedector



Article	Unit	Order-Code	Price (€)
Guard Column	Piece	GUARD	75.00
Guard Column	Pair	GUARD2	140.00

Cardridge Holder for Precolumns with a length of 10 or 20 mm
Order Code K001, Price € 128.00

If you need precolumns of 5mm length additionally the following parts are necessary:

Article	Unit	Order-Code	Price (€)
Distance piece 5 mm	Piece	K0014	15.00
Capillary end fitting ID 0.12 mm (length 100 and 250 mm)	Piece	K0010 (100 mm) K0012 (250 mm)	38.00
Capillary end fitting ID 0.25 mm (length 100 and 250 mm)	Piece	K0015 (100 mm) K0013 (250 mm)	30.50 30.50

SPECIAL COLUMNS

ENVIRONMENTAL ANALYSIS

With the HPLC special columns offered in this catalogue the listed classes of substances can be analyzed according to EPA and DIN regulations, respectively.

UltraSep ES PAH (Polycyclic Aromatic Hydrocarbons)

Our columns especially developed for the determination of **polycyclic aromatic hydrocarbons** reveal simple separation conditions, an excellent resolution (base line separation) of all compounds listed by EPA and DIN as well as short times of analysis. The separation succeeds already with a **simple water/acetonitrile gradient**. Therefore, combined gradients or even ternary gradients are not necessary. Furthermore, a perfect **wave length switching** (fluorescence detection) is possible. Internal standards (e. g. Anthracene-d_{10}, Dibenz(a,h)anthracene-d_{14} or 2-Methylchrysene can be positioned within the chromatogram without difficulties. The often tested columns yield very reproducible results with constant retention times for more than **3000 injections**. They work without problems in the temperature range of 20 – 40 °C.

In the EU it is necessary to control additionally to the PAHs of EPA (Environmental protection Agency, USA) the presence of PAHs from the SFC (Scientific Committee on Food) list (p. E16).

Using precolumns, a contamination of the analytical columns by the samples can be avoided. We recommend our PAH Precolumn Cartridges.

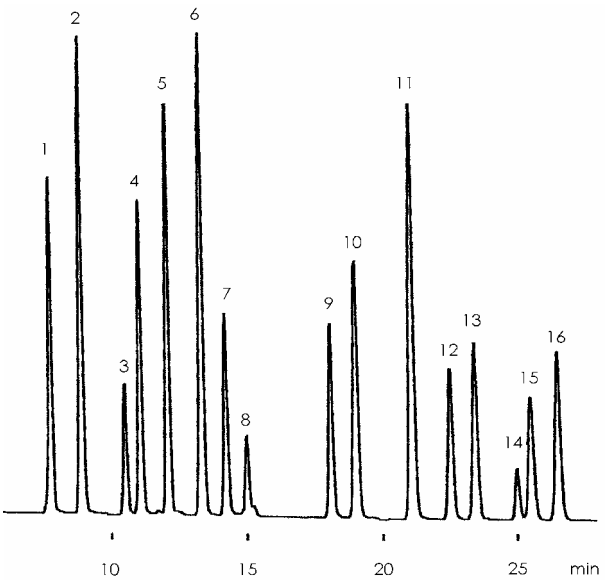
For optimal gradient separations not only the increase of the gradient, but also the adjusted volume flow is of importance. We have optimized the separation conditions for you. The parameters specified in the reference chromatograms are related to high pressure gradients. If you are working with a low pressure gradient, the conditions have to be adapted because of the greater dwell time.

In the case that you can only work isocratically, keep in mind the large requirements of analysis time if higher aromatics are present. We recommend for this case a step gradient, which allows the separation of 2 x 8 = 16 aromatics within approximately 30 minutes: (I) Acetonitrile/water = 55/45, (II) Acetonitrile/water = 90/10.

COLUMNS and **PRICES** (€/piece)

	Column size (mm)					
	250 x 4 200 x 4	**250 x 3*)** 200 x 3	250 x 2 200 x 2	150 x 4 125 x 4	150 x 3 125 x 3	150 x 2 125 x 2
UltraSep ES PAH	350.00	335.00	360.00	330.00	315.00	340.00
	Dimensions (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges	170.00	170.00	170.00	(Price per pack = 5 pieces)		
					125 x 3	
UltraSep ES PAH SUPER					380.00	
	Size (mm)					
	10 x 3	5 x 3				
Precolumn cartridges	170.00	170.00	(Price per pack = 5 pieces)			
	Column size (mm)					
	60 x 4	60 x 3	60 x 2	40 x 2	33 x 3	
UltraSep ES PAH QC	390.00	380.00	390.00	370.00	360.00	
	Size (mm)					
	5 x 4	5 x 3	5 x 2			
Precolumn cartridges QC	170.00	170.00	170.00	(Price per pack=5 pieces)		

*) This column can be used to obtain the same chromatogram quality as it has been shown with AN by a linear water/methanol gradient. Starting pressure 130 bar, analysis time 25 min. See page E-16

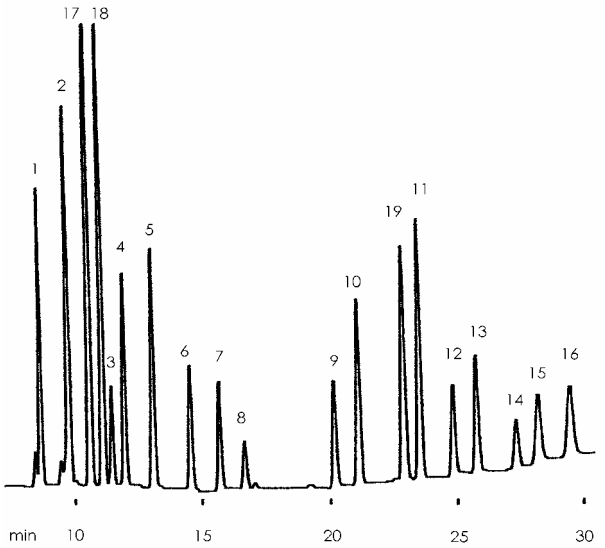


Packing:	UltraSep ES PAH
Column:	125 x 2 mm
Detection:	UV 254 nm
Eluent:	Water/acetonitrile (AN)
Gradient:	Start 35% AN
	25th min 100% AN
Flow rate:	0.4 ml/min
Temperature:	Room temperature
Sample:	Supelco PAH Mix 610-M

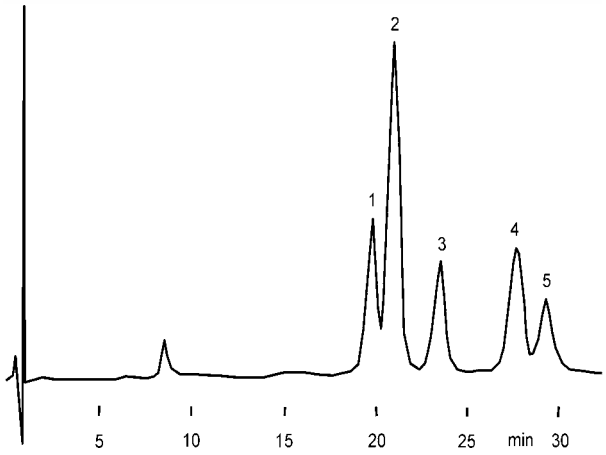
Extended EPA List

1	Naphthalene	9	Benz(a)anthracene
2	Acenaphthylene	10	Chrysene
17*	1-Methylnaphthalene	19*	Benzo(e)pyrene
18*	2-Methylnaphthalene	11	Benzo(b)fluoranthene
3	Acenaphthene	12	Benzo(k)fluoranthene
4	Fluorene	13	Benzo(a)pyrene
5	Phenanthrene	14	Dibenz(a,h)anthracene
6	Anthracene	15	Benzo(g,h,i)perylene
7	Fluoranthene	16	Indeno(1,2,3-c,d)pyrene
8	Pyrene		

* enclosed in the extended EPA list only



Packing:	UltraSep ES PAH
Column:	250 x 3 mm
Detection:	UV 254 nm
Eluent:	Water/acetonitrile (AN)
Gradient:	Start 40% AN
	25th min 100% AN
Flow rate:	0.9 ml/min
Temperature:	Room temperature
Sample :	Supelco PAH Mix 610-M
	(EPA-Aromatics)
	+ 3 additional PAH



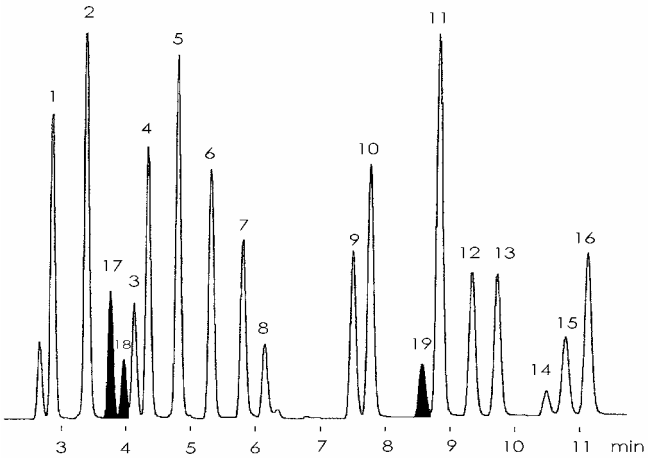
Separation of the Isomeres of
Monohydroxyphenanthrene

1 3-Hydroxyphenanthrene
2 2-Hydroxyphenanthrene
3 9-Hydroxyphenanthrene
4 1-Hydroxyphenanthrene
5 4-Hydroxyphenanthrene

Packing:	UltraSep ES PAH
Column:	250 x 3 mm
Detection:	UV 254 nm
Eluent:	Water/acetonitrile (65/35)
Flow rate:	1 ml/min
Temperature:	20 °C
Sample :	Mixture of isomeresh

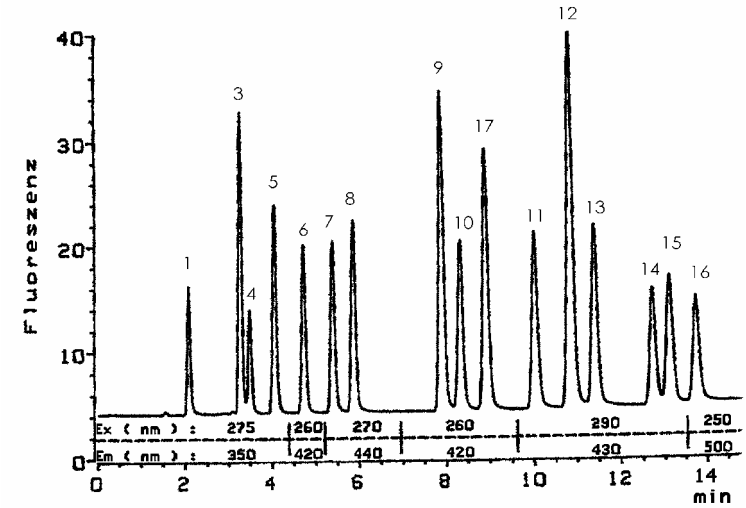
UltraSep ES PAH Quick and **UltraSep ES PAH SUPER**

The top quality packing UltraSep ES PAH is available also with a particle diameter of 4 µm. Because of that the separation columns reach an effectivity of >100000 TP/m and the duration of analysis can be shortened to one third without loss of quality. One advantage of the **Quick Columns** is a rapid adjustment of the equilibrium after each analysis. Therefore, chromatographical runs can follow immediately after each other.



1-16 PAHs accord. to EPA standard
17 1-Methylnaphthalene
18 2-Methylnaphthalene
19 Benzo(e)pyrene

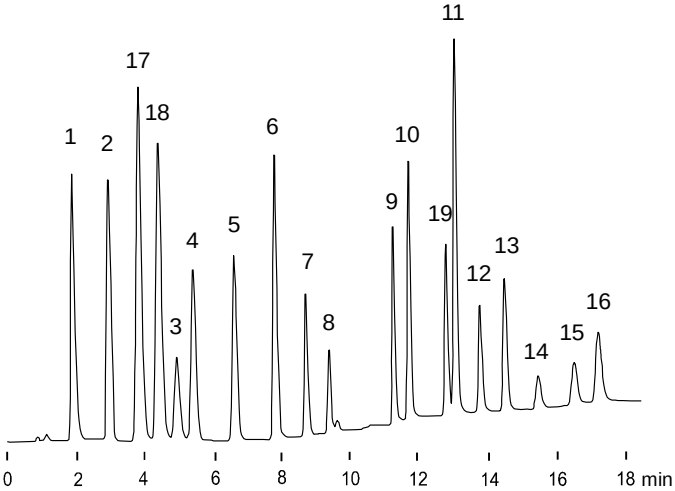
Packing:	UltraSep ES PAH QC
Column:	33x 3 mm
Detection:	UV 254 nm
Eluent:	Water/acetonitrile (AN)
Gradient:	Start 40% AN 11th min 100% AN
Flow rate:	0.7 ml/min
Temperature:	30°C



1, 3-16 PAHs accord. to EPA standard
17 2-Methylchrysene (internal standard)
(2) Acenaphthylene is not detected

Packing:	UltraSep ES PAH QC
Column:	60 x 2 mm
Detection:	Fluorescence, wave length program
Eluent:	Water/acetonitrile (AN)
Gradient:	Start 50% AN 15th min 100% AN
Flow rate:	0.5 ml/min

Chromatogram of a customer
Landesumweltamt Düsseldorf

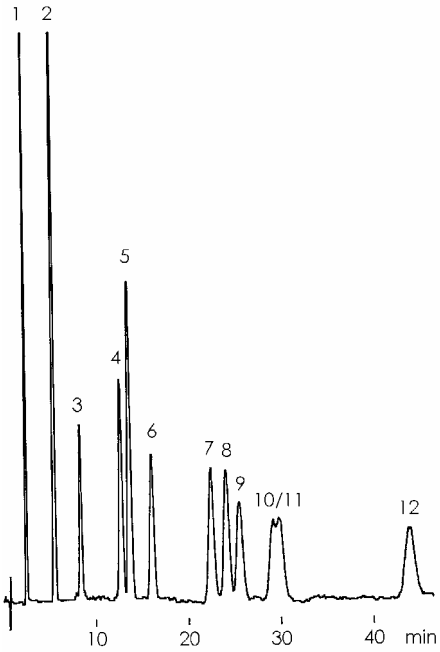


1-16 PAHs accord. to EPA standard
17 1-Methylnaphthalene
18 2-Methylnaphthalene
19 Benzo(e)pyrene

Packing:	UltraSep ES PAH SUPER
Column:	125x 3 mm
Detection:	UV 254 nm
Eluent:	Water/acetonitrile (AN)
Gradient:	Start 55% AN 5th min 55% AN 12th min 100% AN
Flow rate:	0.7 ml/min
Temperature:	25°C

UltraSep ES ALD (Aldehydes and Ketones)

Aldehydes and ketones of ecological relevance, determined as dinitrophenylhydrazones



1	DNPH
2	Formaldehyde
3	Acetaldehyde
4	Acetone
5	Acroleine
6	Propionaldehyde
7	Crotonaldehyde
8	Methylethylketone
9	Cyclopentanon
10/11	n-Butyraldehyde/ iso-Butyraldehyde
12	Benzaldehyde

Packing:	UltraSep ES ALD
Column:	100 x 3 mm
Detection:	UV 350 nm
Eluent:	Water/acetonitrile (65/35)
Flow rate:	0.8ml/min
Temperature:	30 °C

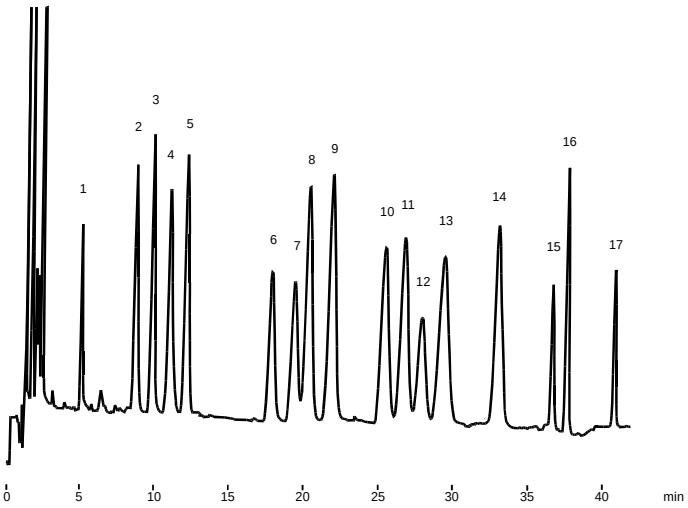
UltraSep ES ALD FF (Insulating Oils)

With these columns furfuryl alcohol, furfurol (furfural) and other aldehydes (degradation products in **insulating oils**) can be determined (according to EN 61198)) without derivatization.

COLUMNS and **PRICES** (€/piece)		
	Column size	
	100 x 3	100 x 2
UltraSep ES ALD	210.00	220.00
UltraSep ES ALD FF	210.00	220.00
	Size (mm)	
	10 x 3	10 x 2
Precolumn cartridges (Price per pack=5 pieces)	170.00	170.00

UltraSep ES PEST (Pesticides)

This packing is characterized by a high selectivity and good separation efficiency. For example, the separation of all the 17 **plant protection products** according to DIN 38407 F12 can be carried out with an acetonitrile/water gradient without problems within a short time. The smallest α-values are 1.05 ± 0.01.



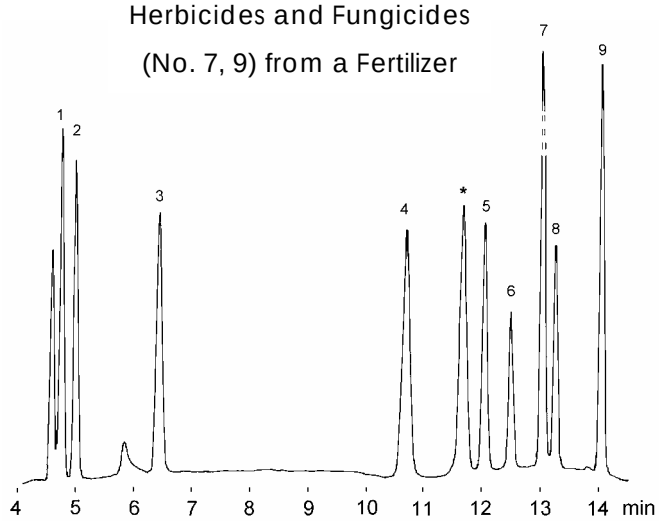
Sequence of 17 plant protection products according to DIN 38407 F12 on UltraSep ES PEST

1	Desethylatrazine	10	Isoproturon
2	Hexazinon	11	Metobromuron
3	Metoxuron	12	Diuron
4	Simazine	13	Metazachlor
5	Cyanazine	14	Sebuthylazine
6	Methabenzthiazuron	15	Terbuthylazine
7	Atrazine	16	Linuron
8	Chlortoluron	17	Metolachlor
9	Monolinuron		

Packing:	UltraSep ES PEST
Column:	250 x 3 mm
Detection:	UV 230 nm
Eluent:	water/acetonitrile)
Gradient:	Start 26% AN
	27th min 26% AN
	40th min 50% AN
	45th min 100% AN
Flow rate:	0.85 ml/min
Temperature:	30°C

Herbicides and Fungicides

(No. 7, 9) from a Fertilizer



Extension of the pesticide range with UltraSep ES PHARM (see page B-2)

Chromatogram of a Customer

Biologische Bundesanstalt für Land- und Forstwirtschaft Berlin

1	Metamitron	6	Phenmedipham
2	Chloridazon	7	Azoxystrobin
3	Carbetamid	8	Ethofumesat
4	Isoproturon	9	Kresoxim-methyl
5	Dimefuron	*	Unknown compound

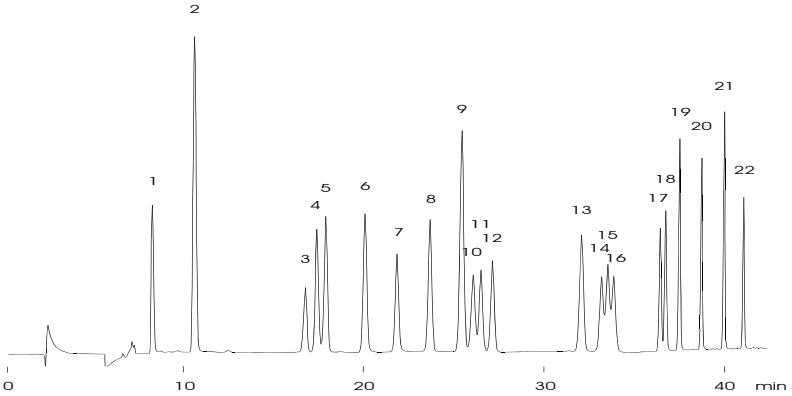
Packing:	UltraSep ES PHARM RP8E 5 µm
Column:	125 x 3 mm
Detection:	UV 230 nm
Eluent:	Water/acetonitrile gradient
Flow rate:	1.0 ml/min
Temperature:	30°C

COLUMNS and **PRICES** (€/piece)

	Column size (mm)		
	250 x 4	250 x 3	250 x 2
UltraSep ES PEST	420.00	410.00	420.00
	Size (mm)		
	10 x 4	10 x 3	10 x 2
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00

UltraSep ES PHENOXYCARB (Herbicides)

This packing is suitable for the determination of herbicides of the phenoxycarbonic acid type in ecological relevant samples.



Packing:	UltraSep ES PHENOXYCARB
Column:	250 x 2 mm
Detection:	UV 224 nm
Eluent:	A: $H_2O+H_2SO_4$ or TFA, pH 2.5 B: acetonitrile
Gradient:	Start 0% B 2nd min 20% B 30th min 35% B 45th min 100% B 50th min stop
Flow rate:	0.3 ml/min
Temperature:	60 °C

Chromatogram of a customer
Berliner Wasserbetriebe
(H. Böhme)

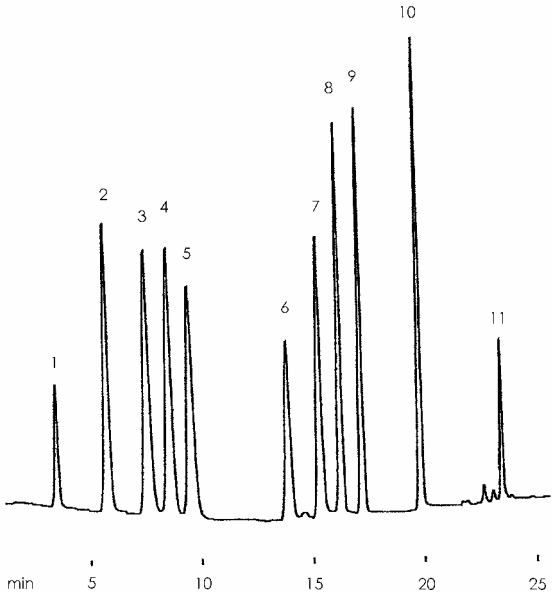
1 Clopyralid
2 Quinmerac
3 Nicosulfuron
4 Dicamba
5 Fluroxypyr
6 Mesotrion
7 Sulcotrion
8 Bentazon
9 Bromoxynil
10 DNOC (4,6-Dinitro-o-cresol)
11 2,4-D (2,4-Dichlorophenoxyacetic acid)
12 MCPA (4-Chloro-o-tolyloxyacetic acid)
13 Ioxynil
14 Dichlorprop
15 Mecoprop
16 2,4,5-T (2,4,5-Trichlorophenoxyacetic acid)
17 2,4-DB [4-(2,4-Dichlorophenoxy)butyric acid]
18 MCPB [4-(4-Chloro-o-tolyloxy)butyric acid]
19 Fluazifop-p
20 Fenoxyprop-p
21 Haloxyfop
22 Dinoterb

COLUMNS and **PRICES** (€/piece)

	Column size					
	250 x 4 200 x 4	250 x 3 200 x 3	250 x 2 200 x 2	150 x 4 125 x 4	150 x 3 125 x 3	150 x 2 125 x 2
UltraSep ES PHENOXYCARB	360.00	340.00	355.00	295.00	280.00	290.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			

UltraSep ES PHEN (Phenols)

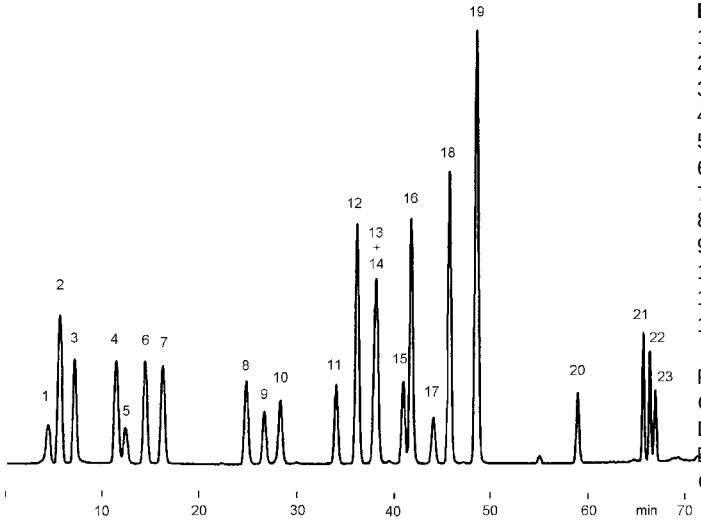
This packing allows the separation of all the heterosubstituted EPA-phenols without interference by phthalates. Furthermore, polyhydric phenols and BTXE-phenols can be separated.



EPA-Phenols

1	Phenol	7	4,6-Dinitro-o-cresol
2	4-Nitrophenol	8	4-Chloro-m-cresol
3	2,4-Dinitrophenol	9	2,4-Dichlorophenol
4	2-Chlorophenol	10	2,4,6-Trichlorophenol
5	2-Nitrophenol	11	Pentachlorophenol
6	2,4-Dimethylphenol		

Packing:	UltraSep ES PHEN
Column:	125 x 3 mm
Detection:	UV 280 nm
Eluent:	Water/methanol; addition of 1% acetic acid to each component
Gradient:	Start 25% Methanol 10th min 35% Methanol 25th min 100% Methanol
Flow rate:	0.85 ml/min
Temperature:	35 °C



BTXE-Phenoles

1	Hydroquinone	13	4-Ethylphenol
2	Resorcinol	14	2,5-Xylenol
3	Pyrocatechol	15	2-Ethylphenol
4	2-Methylpyrocatechol	16	α-Naphthol
5	4-Methoxyphenol	17	Benzene
6	Phenol	18	β-Naphthol
7	Guaiacol	19	2,3,5-Trimethyl-phenol
8	p-Cresol	20	Toluene
9	o-Cresol	21	Ethylbenzene
10	2-Ethoxyphenol	22	o-Xylene
11	3,4-Xylenol	23	p-Xylene
12	3,5-Xylenol		

Packing:	UltraSep ES PHEN
Column:	250 x 3 mm
Detection:	UV 212 nm
Eluent:	Water/acetonitrile (AN)
Gradient:	Start 22% AN 5th min 23% AN 48th min 45% AN 56th min 64% AN
Flow rate:	0.4 ml/min
Temperature:	25°C

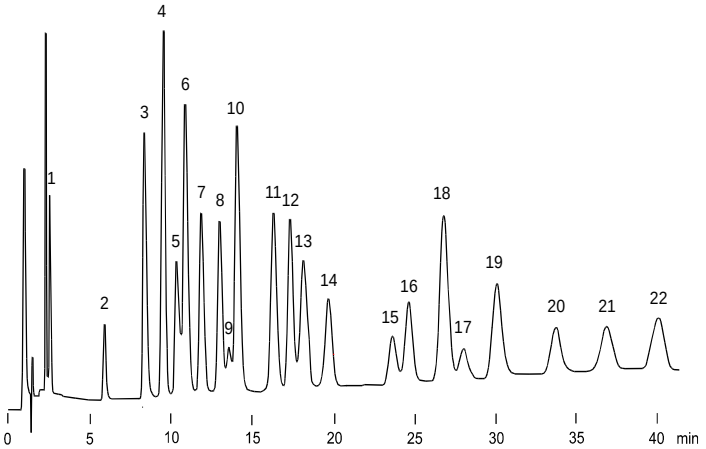
Chromatogram of a customer
Dr. H. Zobel

COLUMNS and **PRICES** (€/piece)

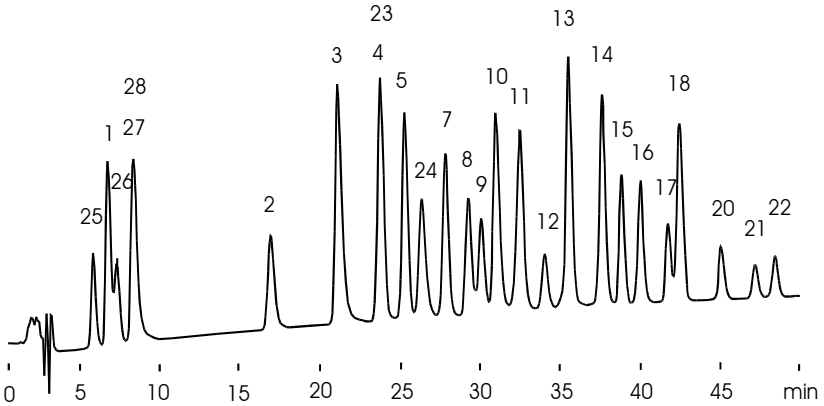
	Column size (mm)					
	250 x 4	250 x 3	250 x 2	125 x 4	125 x 3	125 x 2
UltraSep ES PHEN	220.00	220.00	270.00	195.00	195.00	215.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			

UltraSep ES EX (Explosives)

Explosives as well as chemical weapons, especially in former military training areas, are relevant environmental contaminations. Their recognition and identification by HPLC requires selective packings, which are inert against polar compounds and allow to separate the main explosives sharply from each other. Furthermore it should be possible to identify the most important accompanying and degradation products. These conditions are fullfilled by UltraSep ES EX 424/2, even at isocratic conditions.



Column: 250 x 3 mm
Detection: UV 254 nm
Eluent: Methanol + 0.8Vol.-% n-propanol/water 40/60
Flow rate: 0.7 ml/min
Temperature: 38°C



Packing: UltraSep ES EX 424/2
Column: 250 x 3 mm
Detection: UV 254 nm
Eluent: Water/methanol
Gradient: Start: 35.00% methanol
50th min 66,25% methanol
Flow rate: 0.4 ml/min
Temperature: 23°C

Chromatogram of a customer *Dresdner Grundwasser-forschungszentrum, (Frau Dipl.- Chem. L. Schmalz)*

Nr.	Explosives/accompa-nying substances	Abbre-viation	EPA 8330
1	Octogen *	HMX	x
2	Hexogen **	RDX	x
3	1,3,5-Trinitrobenzene	1,3,5-TNB	x
4	2-Amino-6-nitrotoluene	2-A-6-NT	
5	2-Nitro-4-aminotoluene	2-N-4-AT	
6	1,4-Dinitrobenzene	1,4-DNB	
7	2-Amino-4-nitrotoluene	2-A-4-NT	x
8	1,2-Dinitrobenzene	1,2-DNB	
9	Tetryl ***	-	x
10	1,3-Dinitrobenzene	1,3-DNB	x
11	2,4,6-Trinitrotoluene	TNT, Trotyl	x
12	Nitrobenzene	NB	x
13	4-Amino-2,6-dinitrotoluene	4-A-2,6-DNT	x
14	2-Amino-4,6-dinitrotoluene	2-A-4,6-DNT	x
15	3,4-Dinitrotoluene	3,4-DNT	x
16	2,6-Dinitrotoluene	2,6-DNT	x
17	2,3-Dinitrotoluene	2,3-DNT	
18	2,4-Dinitrotoluene	2,4-DNT	x
19	3,5-Dinitrotoluene	3,5-DNT	
20	2-Nitrotoluene	2-NT	x
21	4-Nitrotoluene	4-NT	x
22	3-Nitrotoluene	3-NT	x
23	2-Methylaniline	2-MA	
24	4-Methylaniline	4-MA	
25	2,6-Diaminotoluene	2,6-DAT	
26	2,6-Diamino-4-nitrotoluene	2,6-DA4NT	
27	2,4-Diamino-6-nitrotoluene	2,4-DA6NT	
28	2,4-Diaminotoluene	2,4-DAT	

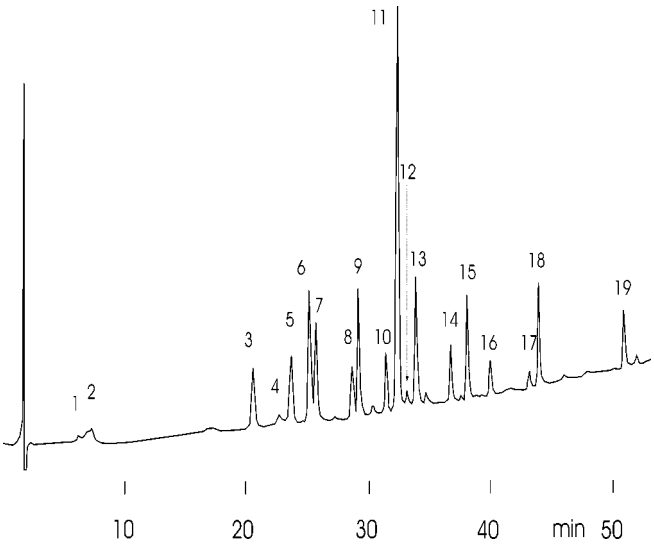
Further names: * 1,3,5,7-Tetranitro-1,3,5,7-tetrazocan
** Hexahydro-1,3,5-trinitro-1,3,5-triazine
*** N-Methyl-N,2,4,6-tetranitroaniline

COLUMNS and **PRICES** (€/piece)

	Column size (mm)					
	250 x 4 200 x 4	250 x 3 200 x 3	250 x 2 200 x 2			
UltraSep ES EX 424/2	340.00	315.00	335.00			
	Size (mm)					
	20 x 4	20 x 3	20 x 2	10 x 4	10 x 3	10 x 2
Precolumn cartridges (Price per pack = 5 pieces)	195.00	195.00	205.00	170.00	170.00	170.00

UltraSep ES AZO (Azo Dyes)

This packing can be used to separate special **azo dyes** according to „Amtliche Sammlung von Untersuchungsverfahren nach §35 LMBG“. The application of these azo dyes is prohibited during the production and treatment of distinct textile consumption goods. The determination is **based on the separation of aromatic amines**, which form at the reductive treatment of the materials.



Packing:	UltraSep ES AZO
Column:	250 x 3 mm
Detection:	UV 240 nm
Eluent:	A: Phosphate buffer pH 6.9
	B: Methanol
Gradient:	Start 15% B
	5th min 15% B
	55th min 80% B
	65th min 80% B
Flow rate:	0.8 ml/min
Temperature:	40 °C

Chromatogram of a customer
Infra Leuna

1	2,4-Diaminoanisol	11	2-Naphthylamine
2	2,4-Toluylenediamine	12	3,3’-Dimethylbenzidine
3	o-Toluidine	13	4-Chloro-o-toluidine
4	Benzidine	14	2,4,5-Trimethylaniline
5	4,4’-Oxydianiline	15	3,3’-Dimethyl-4,4’-diaminodiphenylmethane
6	p-Chloraniline	16	4-Aminodiphenyl
7	2-Amino-4-nitrotoluene	17	3,3’-Dichlorbenzidine
8	p-Cresidine	18	4,4’-Methylene-bis(2-chloroaniline)
9	4,4’-Diaminodiphenylmethane	19	o-Aminoazotoluene
10	4,4’-Thiodianiline		

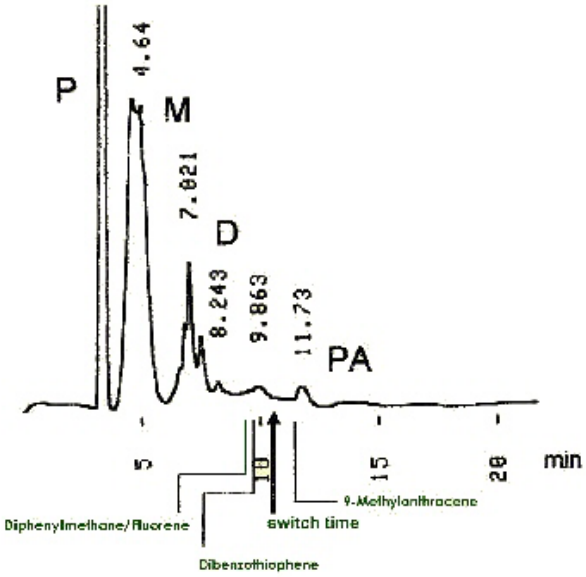
COLUMNS and **PRICES** (€/piece)

	Column size (mm)		
	250 x 4	250 x 3	250 x 2
UltraSep ES AZO	295.00	290.00	290.00
	Size (mm)		
	10 x 4	10 x 3	10 x 2
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00

UltraSep ES AP (Hydrocarbons in Diesel Fuels)

UltraSep ES AP is a special amino packing for the analysis of hydrocarbon classes especially in **diesel fuels**. It is characterized by the following properties:

- Optimal separation of paraffines from aromatics with a resolution of $R_S = 8 – 9$.
- Tetraline eluates always within the family of monoaromatics.
- Diphenylmethane and fluorene eluate clearly before the switch time, which will be calculated with the retention times of dibenzothiophene and 9-methylanthracene.
- Diphenylmethane and fluorene eluate together with or earlier than dibenzothiophene, resp..



P Paraffins
M Monoaromatics
D Diaromatics
PA Polyaromatics

Packing:	UltraSep ES AP
Column:	250 x 4 mm
Detection:	RI
Eluens:	n-Heptane, free of water
Flow rate:	0,6 ml/min
Temperature:	25 °C
Sample:	Diesel fuel

COLUMNS and **PRICES** (€/piece)

	Column size	
	250 x 4	250 x 3
UltraSep ES AP	345.00	330.00
	Size (mm)	
	10 x 4	10 x 3
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00

This column is also qualified without limitation for the determination of FAME (fatty acid methyl esters) in Diesel fuels.

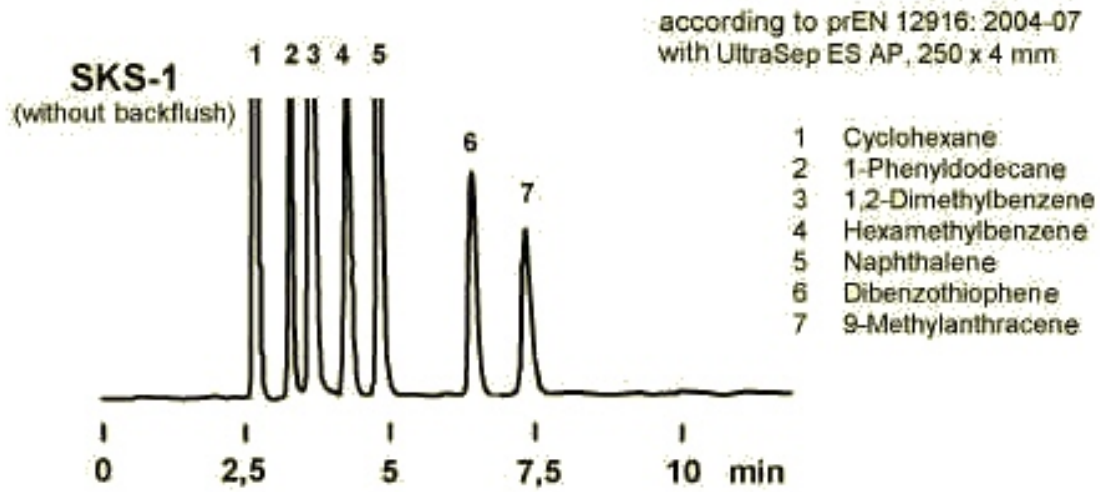
Procedure

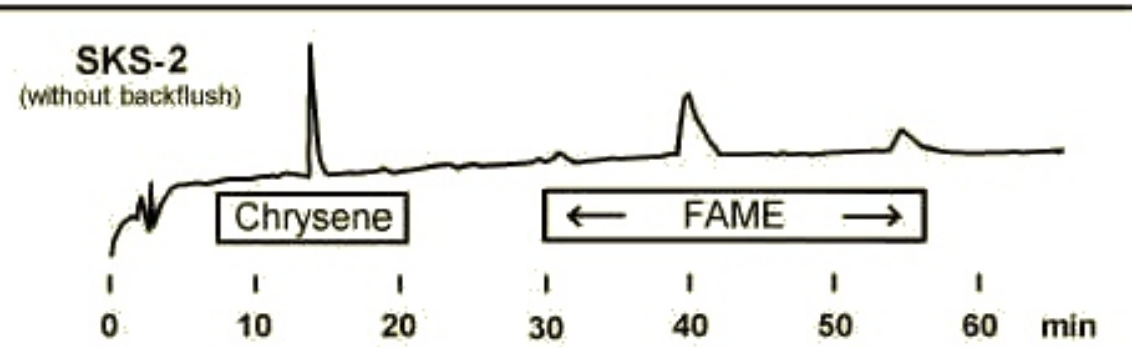
1) SKS-1 (System calibration standard 1) consisting of Cyclohexane, 1-Phenyldodecane, 1,2-Dimethylbenzene, Hexamethylbenzene, Naphthalene, Dibenzothiophene, and 9-Methylanthracene. The components must show base line separation.

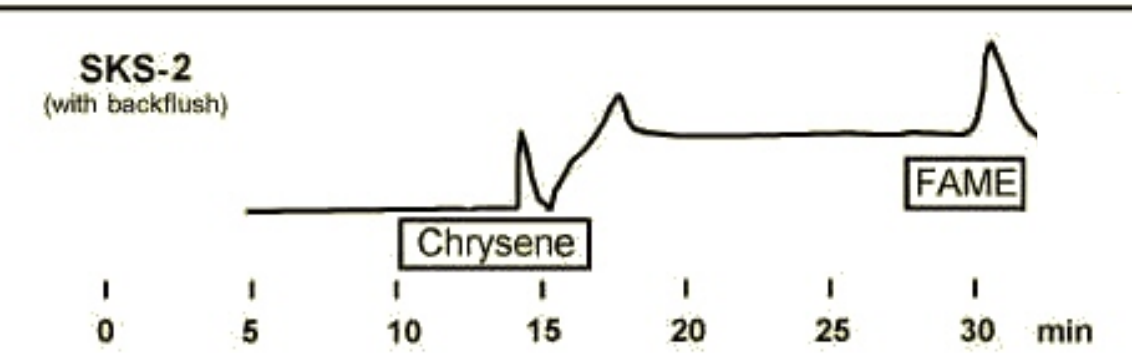
2) SKS-2 (System calibration standard 2) with chrysene und FAME (linolic acid methyl ester, linolenic acid methyl ester, oleic acid methyl ester)

3) Chromatography with backflush after the chrysene peak

(Chromatograms: Dr. G. Liebscher, CHEMTEC Leuna, FB Analytik)

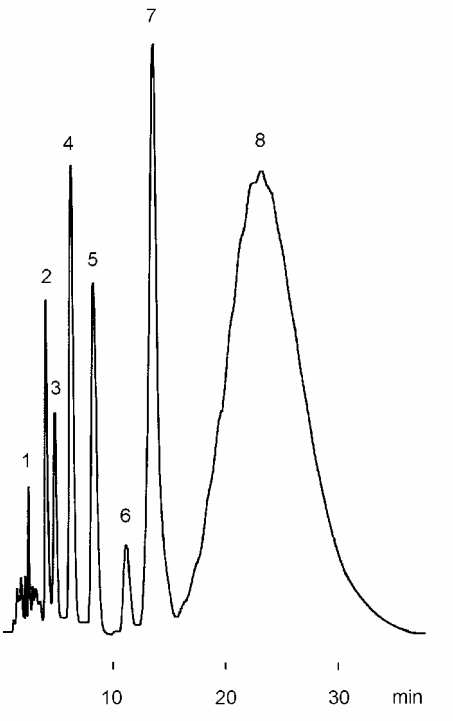






UltraSep ES TENSID
(Alkylbenzene sulfonates, Alkylphenolethoxylates)

Alkylbenzene sulfonates (ABS, LABS, LAS) are the quantitative most important tensides for household and industry. Therefore their determination especially in environmental samples is an important problem. Other tensides relevant to environment are alkylphenolethoxylates. These **nonionic tensides** play only a small part in quantitative relation in comparison to other tensides, but they posses very unfavorable ecological characteristics. Both families can be determined by UltraSep ES TENSID simultaneously with the toxic phenols. The application shows behind of pentachlorophenol the alkylbenzene sulfonates of a sample of Marlon (HÜLS). The isomeres with C10, C11, C12, and C13 alkyl chains give one peak each. After them there appear the ethoxylated phenols as a group peak (C number distribution), fully separated from the phenols they are based on.



1 Chloro- and nitrophenols
2 Pentachlorophenol
3 LAS C10
4 LAS C11
5 LAS C12
6 LAS C13
7 Nonylphenols
8 Nonylphenolethoxylates give a distribution curve

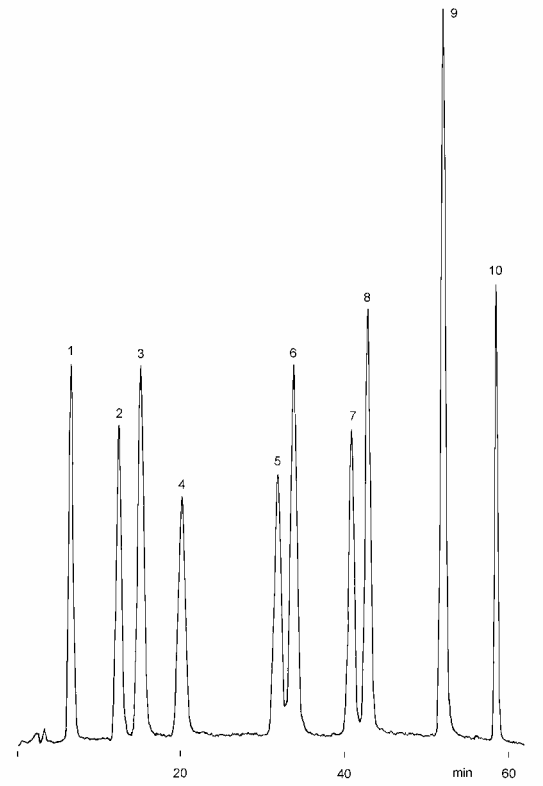
Packing:	UltraSep ES TENSID
Column:	250 x 3 mm
Detection:	UV 220 nm
Eluent:	5 mM Phosphoric acid (pH 4.5)/ Acetonitrile/methanol (47/5/48)
Flow rate:	0.6 ml/min
Temperature:	20 °C

COLUMNS and **PRICES** (€/piece)

	250 x 4	250 x 3	125 x 4	125 x 3
UltraSep ES TENSID	290.00	290.00	260.00	260.00
	Size (mm)			
	10 x 4	10 x 3		
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00		

UltraSep ES AMIN (Amines)

Analyses of room air, water, fitted carpets and other samples require the determination of **aliphatic amines**. After derivatization by 2,4-dinitrofluorobenzene these can be performed with advantage by HPLC.



1 Ethanolamine
2 Methylamine
3 Dimethylamine
4 Ethylamine
5 Propylamine
6 Diethylamine
7 Butylamine
8 sec-Butylamine
9 Pentylamine
10 Hexylamine

Packing:	UltraSep ES AMIN	
Column:	250 x 2 mm	
Detection:	UV 370 nm	
Eluent:	Water/acetonitrile (AN)	
Gradient:	Start	40% AN
	7th min	40% AN
	40th min	60% AN
	65th min	80% AN
Flow rate:	0.2 ml/min	
Temperature:	30°C	

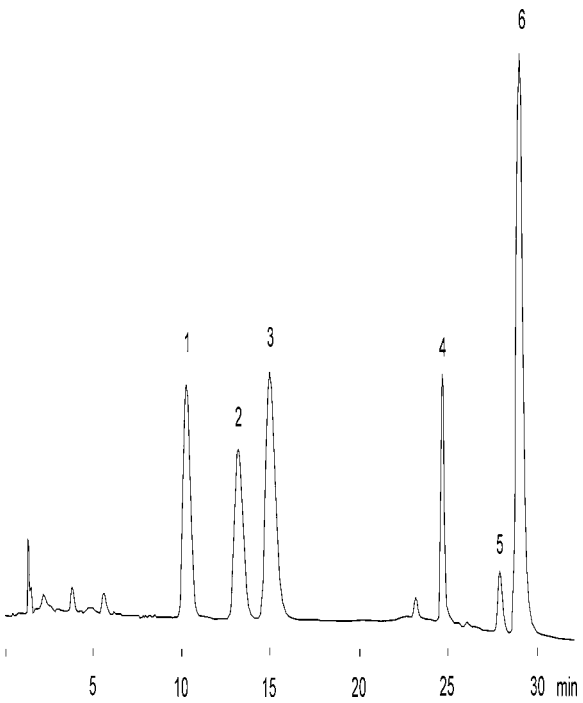
Chromatogram of a customer
Institut Fresenius Gruppe

COLUMNS and **PRICES** (€/piece)

	Column size (mm)					
	250 x 4 200 x 4	250 x 3 200 x 3	250 x 2 200 x 2	150 x 4 125 x 4	150 x 3 125 x 3	150 x 2 125 x 2
UltraSep ES AMIN	350.00	335.00	360.00	330.00	315.00	340.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			

UltraSep ES DIISO (Diisocyanates)

The application of polyurethanes in different branches of industry requires the determination of the ecological relevant **diisocyanates**, which are needed for the production of these plastics.



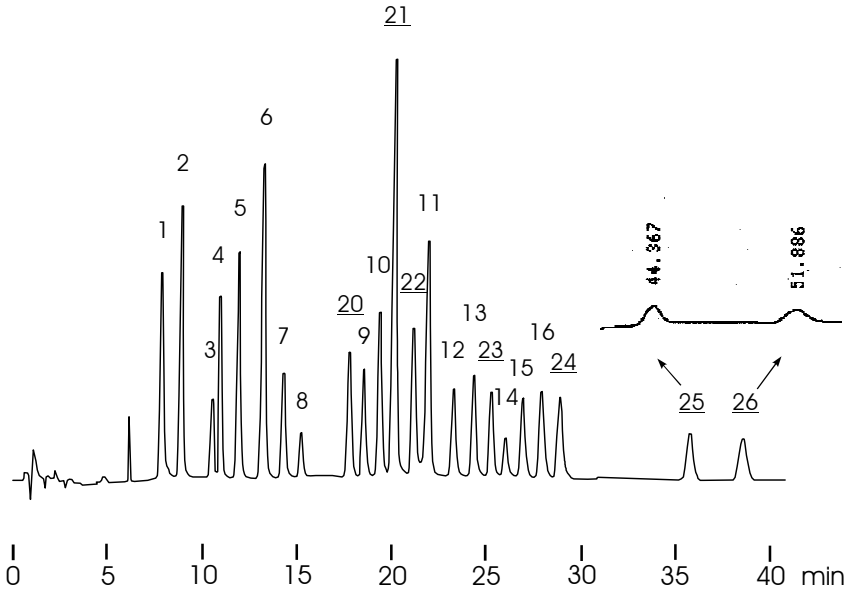
1 2,6-Toluylenediisocyanate (2,6-TDI)
2 Hexamethylene-1,6-diisocyanate (HDI)
3 2,4-Toluylenediisocyanate (2,4-TDI)
4 cis-Isophorondiisocyanate (cis-IPDI)
5 trans-Isophorondiisocyanate (trans-IPDI)
6 4,4'-Methylenediphenylisocyanate (MDI)

Packing:	UltraSep ES DIISO
Column:	250 x 4 mm
Detection:	UV 254 nm
Eluent:	A: 0.1 M ammonia acetate pH 6,2 (with glacial acetic acid)/ 2% acetonitrile B: acetonitrile
Gradient:	Start 27,3% B 17th min 27,3% B 18th min 40% B 30th min 40% B 40th min 27,3% B
Flow rate:	1,65 ml/min
Temperature:	25°C
Derivatized with:	1-(2-pyridyl)piperazine

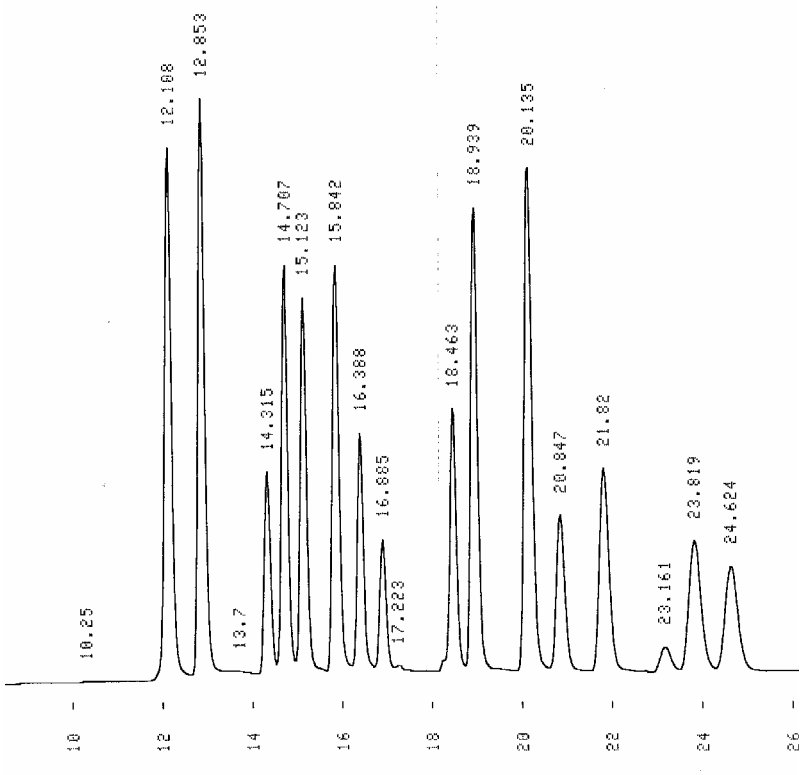
Chromatogram of a customer
Wehrwissenschaftliches Institut Munster

COLUMNS and **PRICES** (in €/piece)

	Column size					
	250 x 4 200 x 4	250 x 3 200 x 3	250 x 2 200 x 2	150 x 4 125 x 4	150 x 3 125 x 3	150 x 2 125 x 2
UltraSep ES DIISO	280.00	275.00	275.00	250.00	245.00	245.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			



Packing: UltraSep ES PAH EFSA
Column: **125 x 3 mm**
Detektion: UV 254 nm
Eluent: water/acetonitrile (AN)*
Gradient: start 40% AN
25. min 100% AN
Flow: 0,6 ml/min
Temperature: 25°C
sample: EPA/EFSA-Aromatics
* at 29. min AN+10%THF; a basic line disturbance due to switching behind compound 24 was suppressed. Above, on the right: Retention times of compounds 25 and 26 without THF-addition). Also a flow gradient behind peak 24 brings advantages.



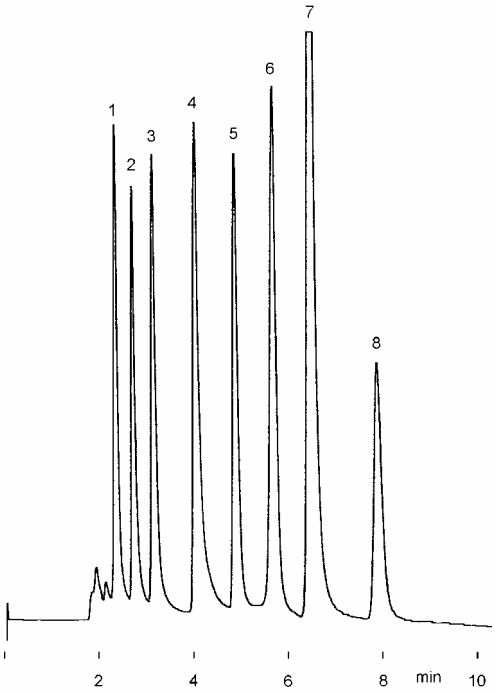
Naphthalene	12.108 min
Acenaphthylene	12,853 min
Acenaphthene	14.315 min
Fluorene	14.707 min
Phenanthrene	15.123 min
Anthracene	15.842 min
Fluoranthene	16.388 min
Pyrene	16.885 min
Benz(a)anthracene	18.463 min
Chrysene	18.939 min
Benzo(b)fluoranthene	20.135 min
Benzo(k)fluoranthene	20.847 min
Benzo(a)pyrene	21.820 min
Dibenz(a,h)anthracene	23.161 min
Benzo(g,h,i)perylene	23.819 min
Indeno(1,2,3-c,d)pyrene	24.624 min

Packing UltraSep ES PAH
Column 250x3mm
Detektion UV 254 nm
Eluent water/methanol (M)
HPLC Gradient Grade
Gradient linear
Flow 0,4 ml/min
Sample Supelco PAH Mix
Pressure at start 130 bar

FOOD STUFF ANALYSIS

UltraSep ESD FS (Fruit Acids)

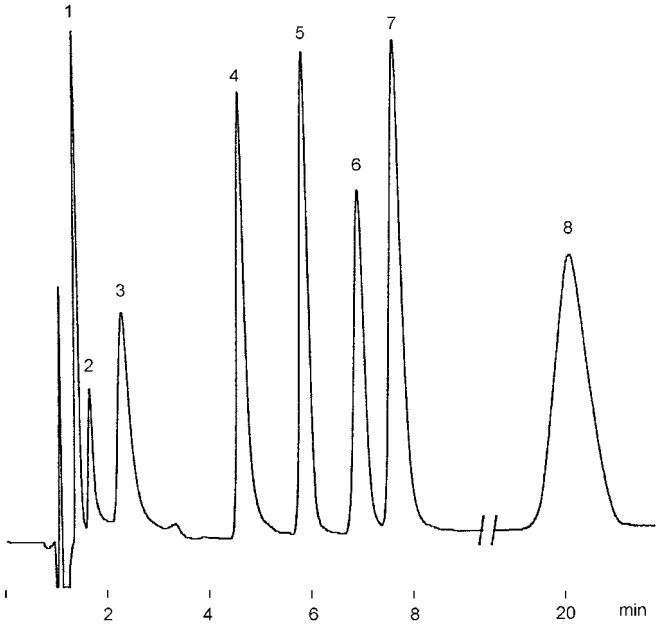
This packing was developed for the separation of small molecular acids, especially of fruit acids in food stuffs and similar materials. In the simplest case it is possible to use the method of ion suppression:



1 Tartaric acid
2 Formic acid
3 Malic acid
4 Lactic acid
5 Acetic acid
6 Citric acid
7 Fumaric acid
8 Succinic acid

Packing:	UltraSep ESD FS, 3 µm
Column:	250 x 3 mm
Detection:	UV 210 nm
Eluent:	10 mM Phosphoric acid, pH 2,9 (NaOH)
Flow rate:	0.60 ml/min
Temperature:	Room temperature

If the mixtures of fruit acids cannot be separated satisfactory in this manner, individual solutions can be achieved by ion pairing chromatography (IPC):



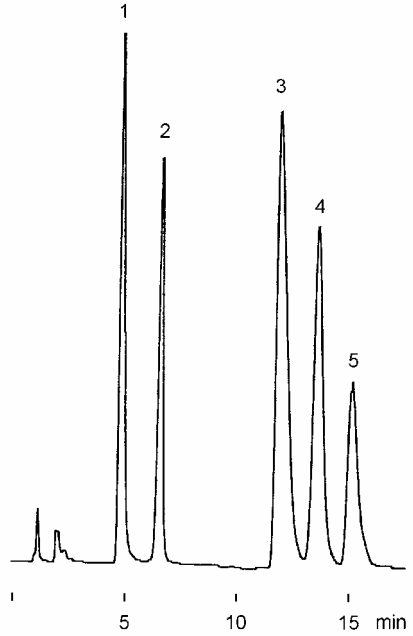
1 Quinic acid
2 Glycolic acid
3 Formic acid
4 Succinic acid
5 Tartaric acid
6 Fumaric acid
7 Oxallic acid
8 Citric acid

Packing:	UltraSep ESD FS, 3 µm
Column:	125 x 3 mm
Detection:	UV 210 nm
Eluent:	Phosphoric acid with ion pairing agent/acetonitrile
Flow rate:	0.55 ml/min
Temperature:	Room temperature

UltraSep ES WAX FS and **UltraSep ES WAXI FS** (Fruit Acids)

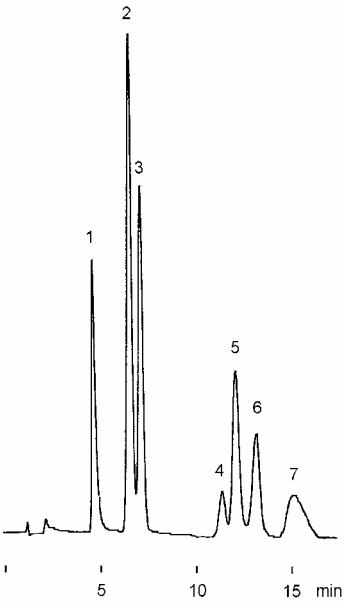
For the case that special mixtures of **mono- and di- or tricarbonic acids** cannot be separated satisfactory by the methodical variants mentioned above, it is possible to determine the monocarbonic acids separately by the ion exchanger UltraSep ES WAX FS. Di- and tricarbonic acids will be removed from the column by back-flushing and, if necessary, separated by UltraSep ESD FS.

The weak imino groups carrying ion exchanger UltraSep ES WAXI FS extends the selectivity of UltraSep ES WAX FS for monocarbonic acids within the pH range of 3.0 – 3.5 (picture at the bottom of the page).



1 Acetic acid
2 Ascorbic acid
3 Lactic acid + glycolic acid
(nearly identical retention times)
4 Quinic acid
5 Formic acid

Packing:	UltraSep ES WAX FS
Column:	250 x 3 mm
Detection:	UV 210 nm
Eluent:	5 mM Phosphoric acid, pH 3,5 (NaOH)
Flow rate:	0.6 ml/min
Temperature:	20 °C

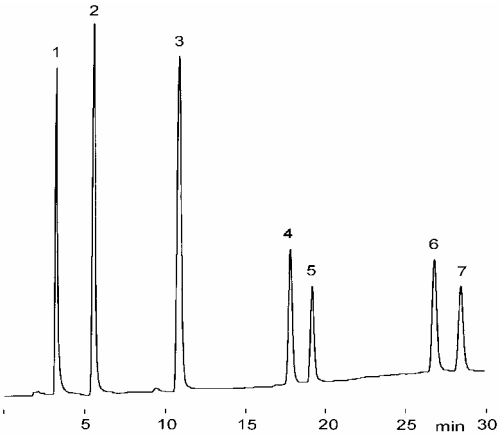


1 Acetic acid
2 Shikimic acid
3 Ascorbic acid
4 Lactic acid
5 Glycolic acid
6 Quinic acid
7 Formic acid

Packing:	UltraSep ES WAXI FS
Column:	250 x 3 mm
Detection:	UV 210 nm
Eluent:	5 mM Phosphoric acid, pH 3.2 (NaOH)
Flow rate:	0.6 ml/min
Temperature:	20 °C

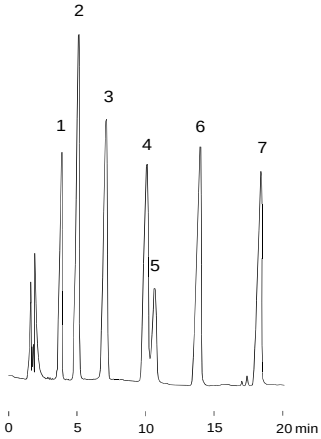
UltraSep ES FS (Fatty Acids)

With this packing the separation of homologeous **fatty acids** by gradient elution is no problem. For the higher fatty acids we recommend UltraSep ES RP18 M500 (see page O-3, prices see O6).



1 Acetic acid
2 Propionic acid
3 n- u. iso-Butyric acid
4 iso-Valeric acid
5 n-Valeric acid
6 iso-Capronic acid
7 n-Capronic acid

Packing:	UltraSep ES FS
Column:	250 x 3 mm
Detection:	UV 210 nm
Eluent:	10 mM Phosphoric acid pH 2,9 (NaOH)/Acetonitrile (AN) Gradient Start 10% AN 20th min 30% AN
Flow rate:	0.55 ml/min
Temperature:	Room temperature



1 Caprinic acid
2 Lauric acid
3 Myristic acid
4 Palmitinic acid
5 Elaidinic acid
6 Stearic acid
7 Arachinic acid

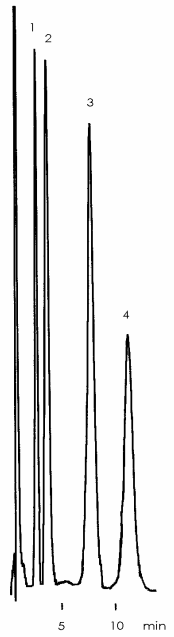
Packing:	UltraSep ES RP18 M500
Column:	250 x 3 mm
Detection:	UV 210 nm
Eluent:	10 mM Phosphoric acid pH 2,9 (NaOH)/ Acetonitrile (AN) Grafient Start 70% AN 30th min 98% AN
Flow rate:	0.55 ml/min
Temperature:	30°C

COLUMNS and **PRICES** (€/piece)

	Column size					
	250 x 4	250 x 3	250 x 2	125 x 4	125 x 3	125 x 2
UltraSep ESD FS	360.00	340.00	355.00	320.00	295.00	290.00
UltraSep ES WAX FS	250.00	225.00	245.00	220.00	195.00	215.00
UltraSep ES WAXI FS	290.00	290.00	290.00	265.00	265.00	265.00
UltraSep ES FS	220.00	220.00	240.00	195.00	195.00	205.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			

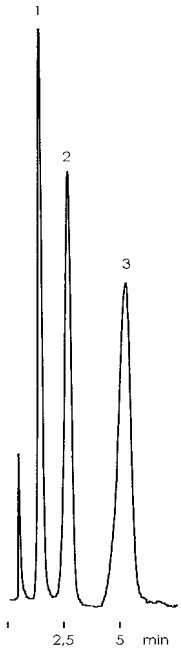
UltraSep ES SACCH (Mono- and Oligosaccharides)

This well-tried packing is based on a special amino phase.



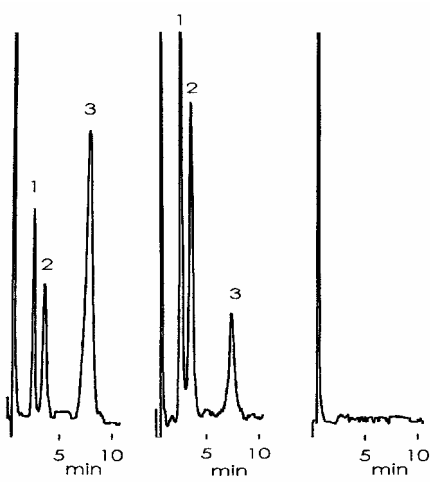
1 Fructose
2 Glucose
3 Saccharose
4 Maltose

Packing:	UltraSep ES SACCH
Column:	125 x 2 mm
Detection:	RI
Eluent:	Water/acetonitrile (15/85)
Flow rate:	0.8 ml/min
Temperature:	Room temperature



1 Glucose
2 Maltose
3 Maltotriose

Packing:	**UltraSep ES SACCH**
Column:	125 x 2 mm
Detection:	RI
Eluent:	Water/acetonitrile (25/75)
Flow rate:	0.8 ml/min
Temperature:	Room temperature



1 Fructose
2 Glucose
3 Saccharose

Packing:	UltraSep ES SACCH
Column:	125 x 2 mm
Detection:	RI
Eluent:	Water/acetonitrile (15/85)
Flow rate:	0.8 ml/min
Temperature:	Room temperature

Samples

left:	Cola from manufacturer 1
middle:	Cola from manufacturer 2
right:	Cola Light

COLUMNS and **PRICES** (€/piece)

	Column size					
	250 x 4	250 x 3	250 x 2	125 x 4	125 x 3	125 x 2
UltraSep ES SACCH	250.00	225.00	245.00	220.00	195.00	215.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			

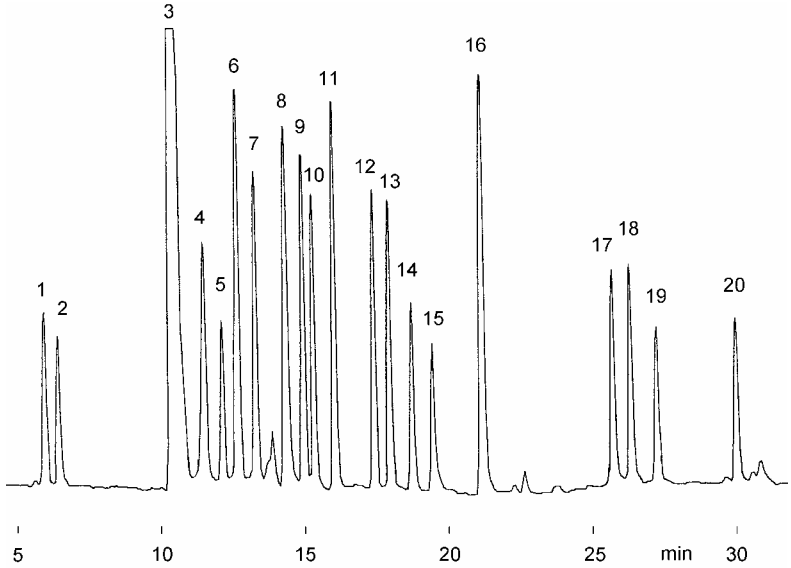
UltraSep ESD AP (Amino Acids, Peptides)

The column is suitable for the separation of derivatives of amino acids and peptides. The common procedures of derivatization are summarized in the following table. If samples are not often analyzed, esp. DABSYLchloride shows advantages.

Comparison of different procedures for derivatization

Procedure	Detection	Limit of detection	Remarks
DABS	Absorption 436 nm	< 1 pmol	Stable derivatives; primary and secondary amino acids can be determined
OPA	Fluorescence 330/440 nm	< 1 pmol	Pro, Hypro and Cys cannot be determined
FMOC	Fluorescence 265/315 nm	< 1 pmol	His and Cys will be detected only in part

DABS = Dabsylchloride (Dimethylaminoazobenzenesulfonylchloride)
OPA = o-Phthalaldehyde
FMOC = 9-Fluorenylmethoxycarbonylchloride



Amino acids as Dabsyl derivatives

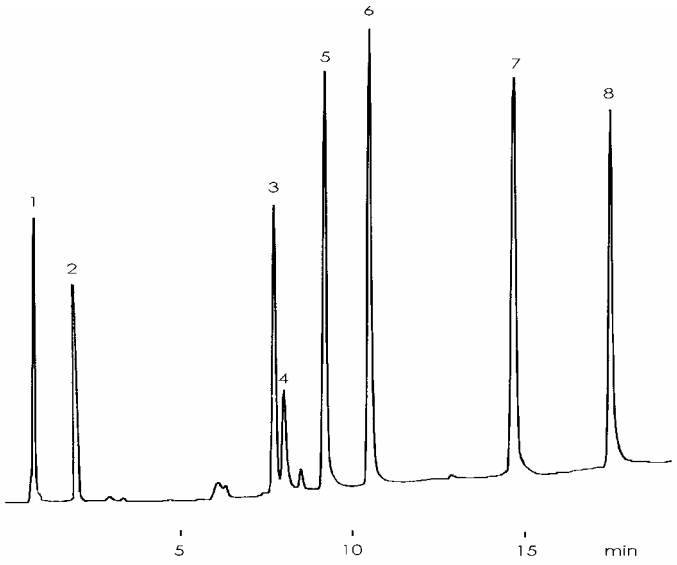
1 Aspartic acid (Asp)
2 Glutamic acide (Glu)
3 DABSOH (Methylorange)
4 Serine (Ser)
5 Threonine (Thr)
6 Glycine (Gly)
7 Alanine (Ala)
8 Arginine (Arg)
9 Proline (Pro)
10 Valine (Val)
11 Methionine (Met)
12 Isoleucine (Ile)
13 Leucine (Leu)
14 Phenylalanine (Phe)
15 unknown
16 Cystine (Cys_2)
17 Ornithine (Orn)
18 Lysine (Lys)
19 Histidine(His)
20 Tyrosine (Tyr)

Packing:	UltraSep ESD AP	Gradient:	Start 4% B
Column:	250 x 3 mm		60th min 100% B
Detection:	436 nm	Flow rate:	0.6 ml/min
Eluent A:	Acetate buffer pH 6,5	Temperature:	45 °C
Eluent B:	Acetonitrile/isopropanol 2/3	Sample diluted 1:1 with acetate buffer	

COLUMNS and **PRICES** (€/piece)

	Column size					
	250 x 4	250 x 3	250 x 2	125 x 4	125 x 3	125 x 2
UltraSep ESD AP	360.00	340.00	355.00	320.00	295.00	290.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			

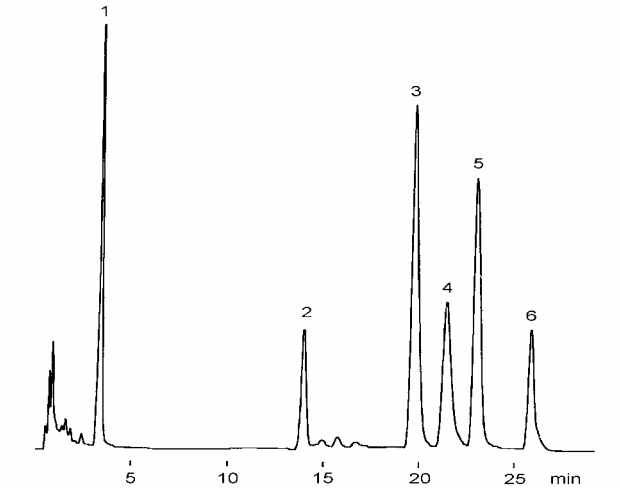
UltraSep ES VIT W (Water Soluble Vitamins)



1 Ascorbic acid (Vitamin C)
2 Nicotinic acid
3 Nicotinamide
4 Folic acid
5 Cyanocobalmine (B12)
6 Riboflavine (B2)
7 Pyridoxine (B6)
8 Thiamine (B1)

Packing:	UltraSep ES VIT W
Column:	125 x 3 mm
Detection:	UV, 280 nm
Eluent:	Water/methanol gradient with addition of ion pairing agent and buffer
Flow rate:	0.5 ml/min
Temperature:	25°C

UltraSep ES VIT F (Fat Soluble Vitamins)



1 Vitamin A (Retinol, Axerophthol)
2 unknown
3 Vitamin D_2
4 Vitamin D_3
5 Vitamin E (α-Tocopherol)
6 Vitamin K1

Packing:	UltraSep ES VIT F
Column:	125 x 2 mm
Detection:	UV 290 nm
Eluent:	Water/acetonitrile (AN)
Gradient:	Start 80% AN 25th min 98% AN
Flow rate:	0.35 ml/min
Temperature:	30 °C

COLUMNS and **PRICES** (€/piece)

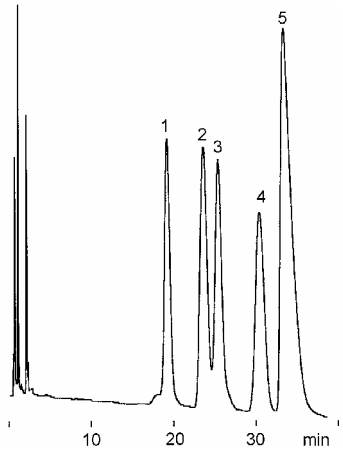
	Column size					
	250 x 4	250 x 3	250 x 2	125 x 4	125 x 3	125 x 2
UltraSep ES VIT W	295.00	275.00	295.00	270.00	250.00	270.00
UltraSep ES VIT F				210.00	210.00	220.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			

UltraSep ES VIT T (Tocopherols)

Two separation columns with UltraSep ES VIT T are offered:

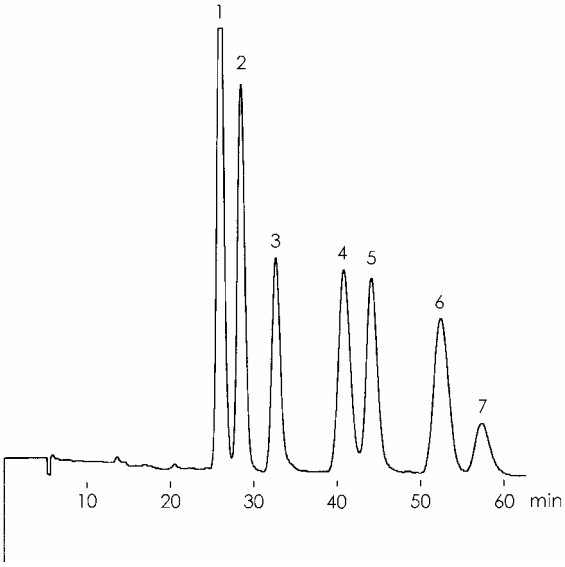
UltraSep ES VIT T1 for the determination of tocopherols including α-tocopherol acetate using RP conditions in aqueous medium and

UltraSep ES VIT T2 for the determination of tocopherols and α -tocopherol acetate under normal phase conditions without water. Up to now for this separation mostly silica columns were used with the known disadvantages (a precise adjustment of the activity is necessary).



1 δ-Tocopherol
2 β-Tocopherol
3 γ-Tocopherol
4 α-Tocopherol
5 α-Tocopherol acetate

Packing:	UltraSep ES VIT T1
Column:	100 x 3 mm
Detection:	UV 290 nm
Eluent:	Water/acetonitrile (15/85)
Flow rate:	0.8 ml/min
Temperature:	30 °C

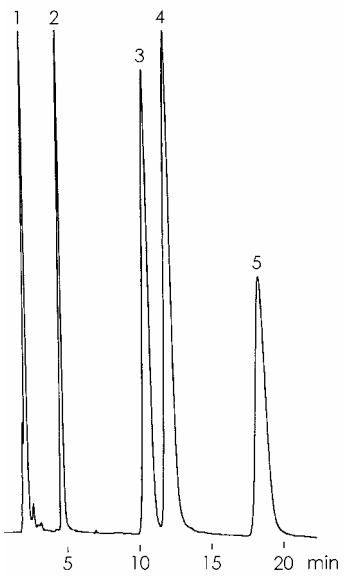


1 Vitamin D_2
2 Vitamin D_3
3 δ-Tocopherol
4 β-Tocopherol
5 γ-Tocopherol
6 α-Tocopherol
7 α-Tocopherol acetate

Packing:	UltraSep ES VIT T1
Column:	100 x 3 mm
Detection:	UV 290 nm
Eluent:	Water/acetonitrile (20/80)
Flow rate:	0.8 ml/min
Temperature:	30 °C

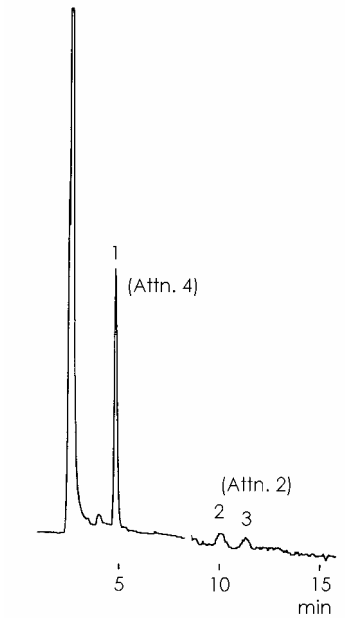
COLUMNS and **PRICES** (€/piece)

	Column size		
	100 x 4	100 x 3	100 x 2
UltraSep ES VIT T1	225.00	225.00	235.00
	Size (mm)		
	10 x 4	10 x 3	10 x 2
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00



1 α-Tocopherol acetate
2 α-Tocopherol
3 β-Tocopherol
4 γ-Tocopherol
5 δ-Tocopherol

Packing:	UltraSep ES VIT T2
Column:	250 x 3 mm
Detection:	UV 290 nm
Eluent:	n-Hexane + 0.35% isopropanol
Flow rate:	0.6 ml/min
Temperature:	Room temperature



1 α-Tocopherol
2 β-Tocopherol
3 γ-Tocopherol

Sample :	Oil from thistle
Packing:	UltraSep ES VIT T2
Column:	250 x 3 mm
Detection:	UV 290 nm
Eluent:	n-Hexane + 0.35% isopropanol
Flow rate:	0.6 ml/min
Temperature:	Room temperature

COLUMNS and **PRICES** (€/piece)

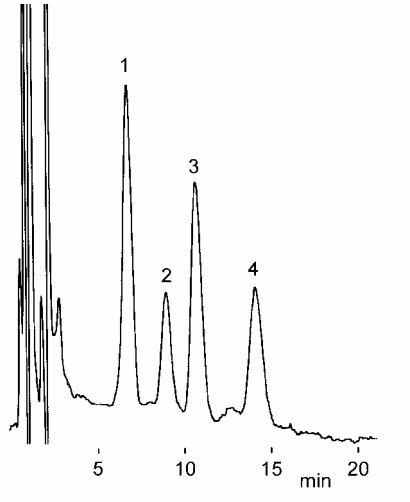
	Column size		
	250 x 4	250 x 3	250 x 2
UltraSep ES VIT T2	240.00	240.00	245.00
	Size (mm)		
	10 x 4	10 x 3	10 x 2
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00

UltraSep ES TOX (Aflatoxins)

The columns quickly separate the mycotoxins Aflatoxin G2, G1, B2, and B1, which are known as liver carcinogenic substances. Especially with UltraSep ES TOX1 the chromatogram is not interfered with eluent peaks. UltraSep ES TOX2 is based on a wide-pore gel for quick analysis.

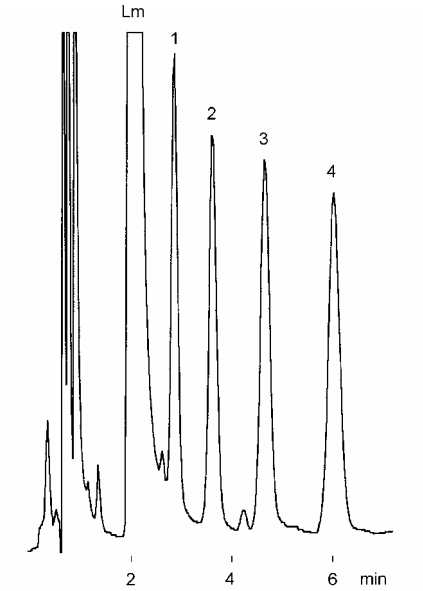
To get a low determination limit of 0.5 ppb, fluorescence detection is used. For this, a post-column derivatization by bromination with pyridinium tribromide (pyridinium bromide perbromide) is necessary.

Best, one works with a CoBrA-Cell (Coring System Diagnostik GmbH, Gernsheim), in which the bromine is generated from KBr electrically.



1 Aflatoxin G2
2 Aflatoxin G1
3 Aflatoxin B2
4 Aflatoxin B1

Packing:	UltraSep ES TOX1
Column:	125 x 3 mm
Detection:	UV 254 nm
Eluent:	Water/methanol (40/60)
Flow rate:	0.6 ml/min
Temperature:	Room temperature
Injektionsvolumen:	20 µl (diluted chloroformic solution)

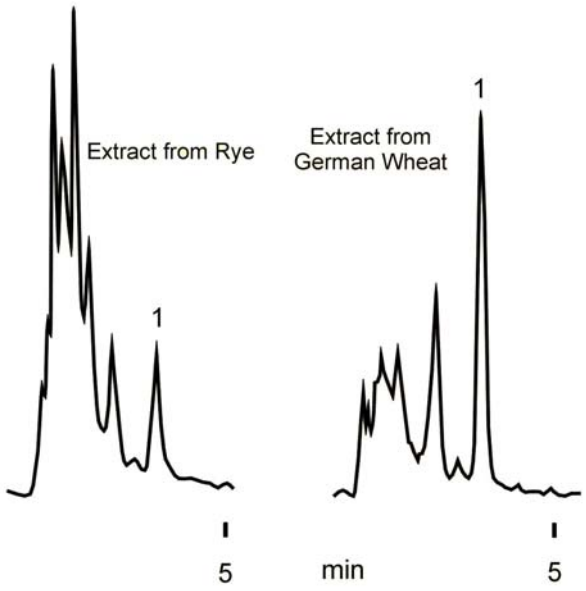


Lm Solvent peak (chloroform)
1 Aflatoxin G2
2 Aflatoxin G1
3 Aflatoxin B2
4 Aflatoxin B1

Packing:	UltraSep ES TOX2
Column:	125 x 3 mm
Detection:	UV 254 nm
Eluent:	Water/methanol (65/35)
Flow rate:	1 ml/min
Temperature:	Room temperature
Injection volume:	3 µl

UltraSep ES TOX O (Ochratoxin A)

This packing is used for the determination of Ochratoxin A, a mycotoxin from Aspergillus und Penicillium species in plants and cereals.



1 Ochratoxin A

Packing:	UltraSep ES TOX O
Column:	125 x 3 mm
Detection:	UV 332 nm
Eluent:	Water/acetonitrile/acetic acid (60/40/1)
Flow rate:	0.6 ml/min
Temperature:	Room temperature
Injection volume :	10 µl

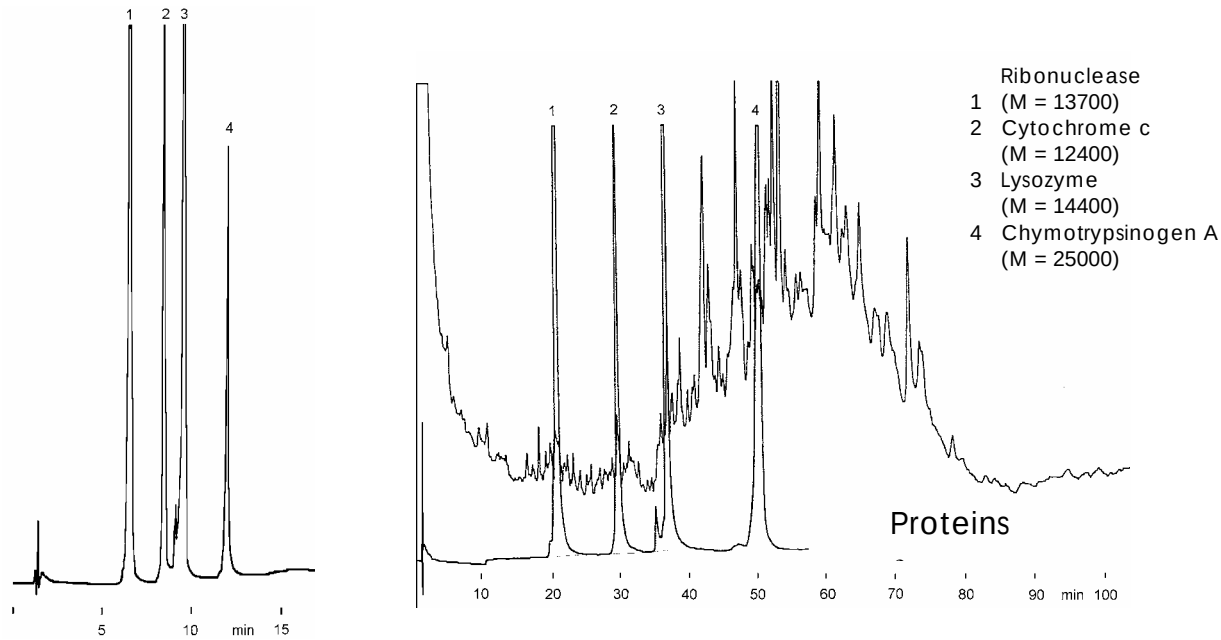
COLUMNS and **PRICES** (€/piece)

	Column size					
	250 x 4	250 x 3	250 x 2	125 x 4	125 x 3	125 x 2
UltraSep ES TOX1 and TOX2	270.00	255.00	270.00	240.00	235.00	240.00
UltraSep ES TOX O				195.00	195.00	230.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00			

BIOCHEMICAL, CLINAL-CHEMICAL AND PHARMACEUTICAL ANALYSIS

UltraSep ESD300 PROT (Proteins)

The packings for the separation of proteins were developed on the basis of a wide-pore material with an average pore diameter of 30 nm. This material allows a high resolution and separations of proteins with high recovery rates.



	Left picture	Right picture
Chromatograms:	Test compounds 1-4	Proteins from Bacillus subtilis with test compounds 1-4 under the same conditions
Packing:	UltraSep ESD300 PROT RP4	UltraSep ESD300 PROT RP18S
Column:	125 x 2 mm	
Detection:	UV 220 nm	
Eluent:	A: 10 mM Trifluoroacetic acid (TFA) in water B : 9 mM TFA in Water/acetonitrile (2/8)	
Gradient:	Start 20% B, 20th min 100% B	Start 20% B, 120th min 100% B, 140th min Stop
Flow rate:	0.25 ml/min	
Temperature	36 °C	

COLUMNS and **PRICES** (€/piece)

	Column size (mm)					
	125 x 4	125 x 3	125 x 2	60 x 4	60 x 3	60 x 2
UltraSep ESD300 PROT RP18	305.00	305.00	305.00	290.00	290.00	295.00
UltraSep ESD300 PROT RP18S*	315.00	315.00	315.00	300.00	300.00	305.00
UltraSep ESD300 PROT RP4	275.00	275.00	275.00	265.00	265.00	265.00
UltraSep ESD300 PROT RP4S*	285.00	285.00	285.00	275.00	275.00	275.00
*endcapped	Size (mm)					
	10 x 4	10 x 3	10 x 2	5 x 4	5 x 3	5 x 2
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00	170.00	170.00	170.00

UltraSep ES PHARM (Pharmaceuticals, especially Bases)

UltraSep ES PHARM and UltraSep ES PHARM SPEZIAL are high selective HPLC-packings, first of all developed to analyze pharmaceutical products, which are often very polar and strong basic. The extreme small content of metallic ions in these packings must be pointed out. With such columns one can work at low buffer concentrations. UltraSep ES PHARM SPEZIAL bears methylalkyl groups and possesses a selectivity different to that of UltraSep ES PHARM. For strongly basic substances UltraSep ES PHARM RP18 SUPER is proved very usefully.

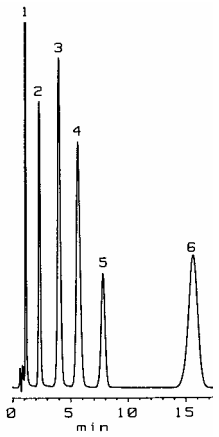
All UltraSep ES PHARM columns are available also with endcapped materials.

In this manner the following main variables can be used for the optimization of a separation:

- Length of the alkyl chain C8/C18
- UltraSep ES PHARM/UltraSep ES PHARM SPEZIAL
- Endcapping
- pH value

For the preparation of buffer solutions it is of advantage to avoid alkali ions. By it the lifespan of the columns will be enlarged. Furthermore there will be no blockage by precipitated salts at high concentrations of organic solvents. Favorable is the adjustment of pH by triethylamine.

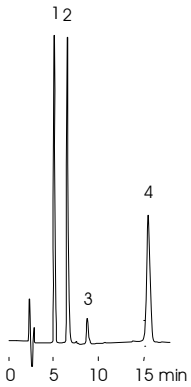
See also **Shield-Carriers,** page O – 3.



1 Hydrochlorothiazide
2 Papaverine
3 Propranolol
4 Diphenhydramine
5 Benzene (as reference peak)
6 Diazepam

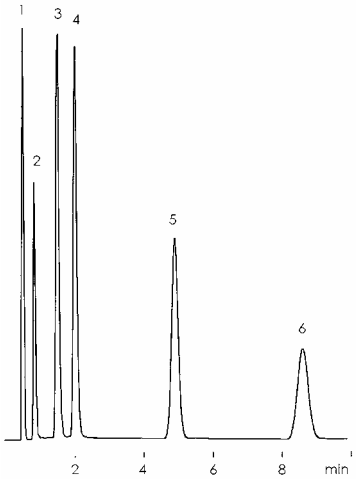
Packing: UltraSep ES PHARM RP8
Column: **100 x 3 mm**
Detection: UV 212 nm
Eluent: 0.015 M Phosphate buffer/acetonitrile (7/3), pH 2.3
Flow rate: 0.9 ml/min
Temp.: Room temperature

Broadband Antibiotics



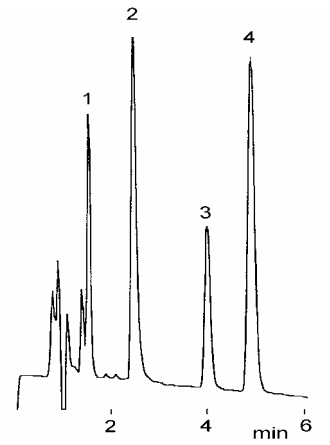
1 Oxytetracycline
2 Tetracycline
3 Contamination in 4
4 Chlorotetracycline

Packing: UltraSep ES PHARM RP18
Column: 150x3mm
Detection: UV 254 nm
Eluent: AN/water (20/80) +0.5% TFA
Flow rate: 0.45 ml/min
Temp.: 30 °C



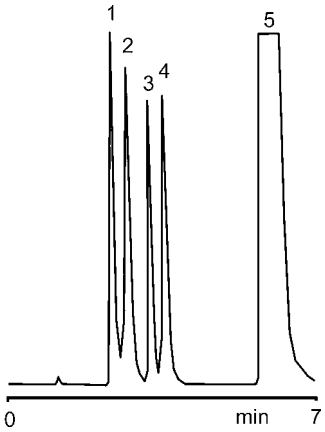
1 Pyridine
2 Coffeine
3 Propranolol
4 Diphenhydramine
5 MPPH
6 Diazepam

Packing: UltraSep ES PHARM SPEZIAL RP8
Column: 125 x 2 mm
Detection: UV 205 nm
Eluent: 0.015 M Phosphate buffer/acetonitrile (7/3), pH 2.3
Flow rate: 0.5 ml/min
Temperature: 30 °C



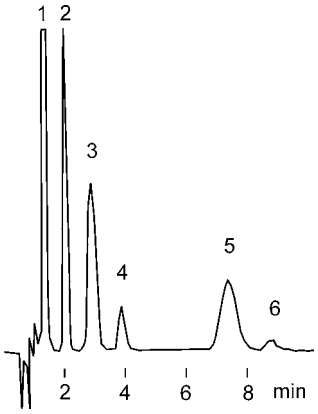
1 Mexiletine
2 Clozapine
(Piperazine ring system)
3 Flecainide
4 Protriptyline

Packing: UltraSep ES PHARM RP8E
Column: 125 x 3 mm
Detection: UV 215 nm
Eluent: 0.015 M Phosphate buffer/acetonitrile (7/3), pH 3.3
Flow rate: 0.6 ml/min
Temperature: 50 °C



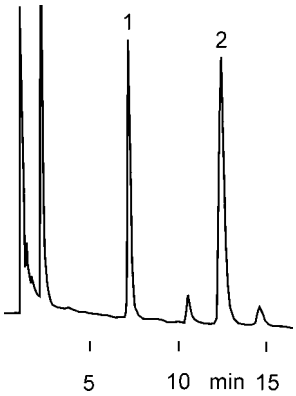
1 4-Hydroxybenzoic acid
2 2-Hydroxyisophthalic acid
3 Phenol
4 4-Hydroxyisophthalic acid
5 Salicylic acid

Sample: **Crude salicylic acid**
Packing: UltraSep ES PHARM RP8
Column: 200 x 3 mm
Detection: UV 230 nm
Eluent: Water/methanol (65/35), pH 2.1
(phosphoric acid)
Flow rate: 1.1 ml/min
Temperature: Room temperature



1, 2, 4, 6 **Peaks from a sample of heroine**
3 Morphine
5 Codeine

Sample of heroine with added morphine and codeine
Packing: UltraSep ES PHARM SPEZIAL RP8
Column: 125 x 2 mm
Detection: UV 205 nm
Eluent: 0.015 M Phosphate buffer/acetonitrile (85/15), pH 7.0
Flow rate: 0.5 ml/min
Temperature: 30 °C

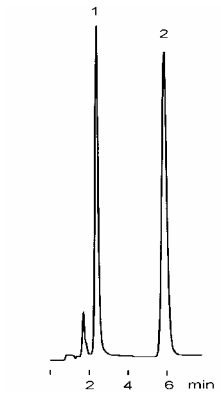


1 Chlordiazepoxide (Tranquilizer)
2 Flumazenil (antidot; benzodiazepine-antagonist)

Packing: UltraSep ESD PHARM RP8 3 µm
Column: 125 x 3 mm
Detection: UV 230 nm
Eluent: 0.050 M Phosphoric acid pH 2.2/acetonitrile (80/20)
Flow rate: 0.6 ml/min
Temperature: Room temperature

1 Histamine (not derivatized)

Packing:	UltraSep ES PHARM RP18
Column:	150 x 3 mm
Detection:	UV 220 nm
Eluent:	0.040 M Phosphoric acid pH 2,0 (adjusted with triethylamine) + 0.005 M dodecyl sulfate
Flow rate:	0.6 ml/min
Temperature:	25 °C



PHARM Packings are also qualified for the chromatography of many biologically important classes of compounds, for instance peptides. An example is the tripeptide **Glutathione** (γ-Glutamyl-cysteinyl-glycine) in the reduced (GSH) and oxidized (GSSG) form.

1 GSH
2 GSSG

Packing:	UltraSep ES PHARM RP18
Column:	125 x 3 mm
Detection:	UV 215 nm
Eluent:	15 mM Phosphoric acid pH 3.3
Flow rate:	0.6 ml/min
Temperature:	30 °C

COLUMNS and **PRICES** (€/piece)

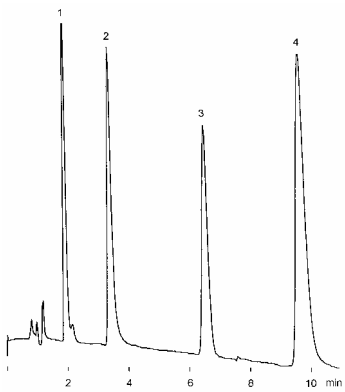
	Column size (mm)					
	250 x 4 200 x 4	250 x 3 200 x 3	250 x 2 200 x 2	150 x 4 125 x 4	150 x 3 125 x 3	150 x 2 125 x 2
UltraSep ES PHARM *)	270.00	265.00	265.00	240.00	235.00	235.00
UltraSep ES PHARM E *)	280.00	275.00	275.00	250.00	245.00	245.00
UltraSep ESD PHARM **)	360.00	340.00	355.00	320.00	295.00	290.00
UltraSep ESD PHARM E **)	370.00	350.00	365.00	330.00	305.00	300.00
UltraSep ES PHARM SPEZIAL *)	295.00	290.00	290.00	265.00	260.00	260.00
UltraSep ES PHARM SPEZIAL E *)	305.00	300.00	300.00	275.00	270.00	270 .00
UltraSep ES PHARM RP18 SUPER	310.00	305.00	305.00	280.00	275.00	275.00
UltraSep ES PHARM RP18 E SUPER	320.00	315.00	315.00	290.00	285.00	285.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges **(Price per pack = 5 pieces)**	170.00	170.00	170.00			

*) RP8 or RP18, 5 µm **) RP8 or RP18, 3 µm; E=Endcapped

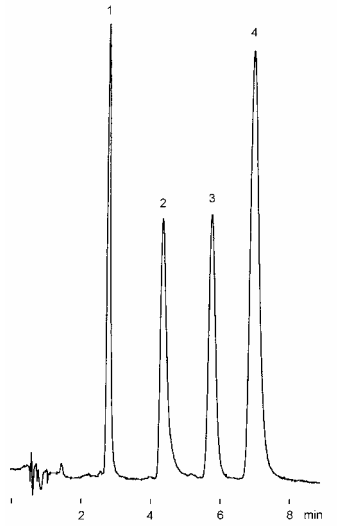
UltraSep ES MLD (Pharmaceuticals, Natural Compounds)

MLD packings (MLD = monolayer dense) are very stable solide supports, known in the literature as "mixed self-assembled monolayers (SAM)". They can be used both for basic and acidic compounds in a relatively wide range of pH and with high selectivity. The buffer concentration can be held less. With organic buffers one can successfully use these columns between pH 2 – 11.

The following picture on the left side demonstrates a separation on UltraSep ES MLD RP18, which was performed already on UltraSep ES PHARM RP8E with 15 mM phosphate buffer as eluent (see the picture on the top of page B-3).



Eluent:
15 mM Phosphoric acid/acetonitrile (70/30); pH 3.3



Eluent:
5 mM Phosphoric acid/acetonitrile (50/50); pH 3.3

1 Mexiletine
2 Clozapine
3 Flecainide
4 Protriptyline

Packing:	UltraSep ES MLD RP18
Column:	125 x 3 mm
Detection:	UV 215 nm
Flow rate:	0.6 ml/min
Temperature:	Room temperature

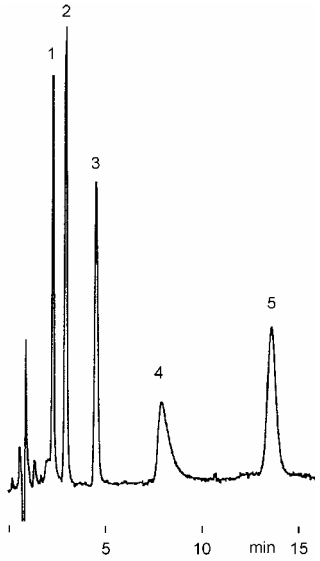
With UltraSep ES MLD RP18 already ≤ 5 mM phosphoric acid is sufficient for delivering the same chromatogram quality (picture on the right side). Furthermore it is important that for obtaining comparable short retention times of basic pharmaceuticals one can work with 50% acetonitrile (instead of 30%).

COLUMN and **PRICES** (€/piece)

	Column size (mm)			
	200 x 3	200 x 2	150 x 3 125 x 3	150 x 2 125 x 2
UltraSep ES MLD (RP8 or RP18 5 µm)	280.00	280.00	255.00	255.00
	Size (mm)			
	10 x 3	10 x 2		
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00		

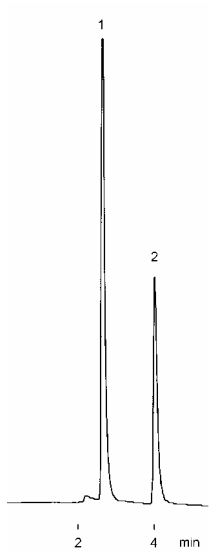
The following application (picture on the left side)represents a chromatogram of basic pharmaceuticals at pH 11, which was obtained after flushing the column with 6000 ml eluent, pH 11, corresponding to a continuous use of approximately 4 weeks. Remarkable is the still very good peak shape especially of perazine dimalonate.

For working with basic eluents the usage of organic buffers and the addition of organic solvents is recommended.



1 Flecainide
2 Clozapine
3 Diphenhydramine
4 Perazindimalonat
5 Amitriptyline

Packing: UltraSep ES MLD RP18
Column: 125 x 3 mm
Detection: UV 215 nm
Eluent: 50 mM 1-Methylpiperidine in water/ acetonitrile (40/60), pH 11
Flow rate: 0.6 ml/min
Temperature: 25 °C

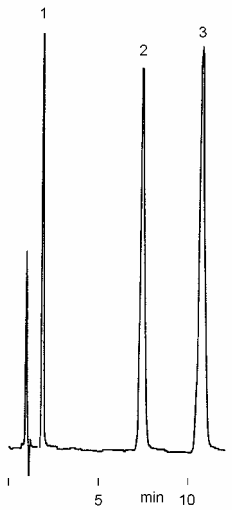


1 Formic acid
2 Acetic acid

Packing: UltraSep ES MLD RP18
Column: 250 x 3 mm
Detection: UV 210 nm
Eluent: 10 mM Phosphoric acid pH 2.9
Flow rate: 0.6 ml/min
Temperature: Room temperature

MLD packing are, as already mentioned, suitable for the separation of organic acids too (picture on the right side).

Using MLD packings it is possible to obtain chromatograms with good peak shapes of very different classes of compounds:



1 Sulfathiazol
2 Chloramphenicol
3 Sulfadimethoxin

Packing: UltraSep ES MLD RP18
Column: 125 x 3 mm
Detection: UV 254 nm
Eluent: Phosphoric acid/acetonitrile (80/20; pH 2.8)
Flow rate: 0.6 ml/min
Temperature: 25 °C

UltraSep ES PFP

UltraSep ES PFP is a pentafluorophenylalkylsilica of the RP8 reversed phase type with outstanding properties. On average it yields the retention times of the RP18 packing type with the following characteristics:

- Compounds which are not separated enough by normal RP18 phases, potentially can be resolved sufficiently using the PFP packing (purity screening).
- PFP is very attractive for pharmaceutically relevant substances, especially for bases, which in principle show a small tailing (picture 1 and 2). Base pairs which are not separated by normal RP packings, can be resolved (picture 3).
- PFP packings behave significantly different in comparison to normal RP packings. For instance in the test mixture of toluene, propyl benzene and pentyl benzene with rising water content the peaks become slender. At a composition of the acetonitrile/water mixture of 1/1 125,000 theoretical plates per m are accomplished already with 4 µm material.
- PFP phases together with the normal RP phases and RP amid phases or also with PEO phases yield an advantageous "selectivity trio" and "selectivity quartet", respectively.

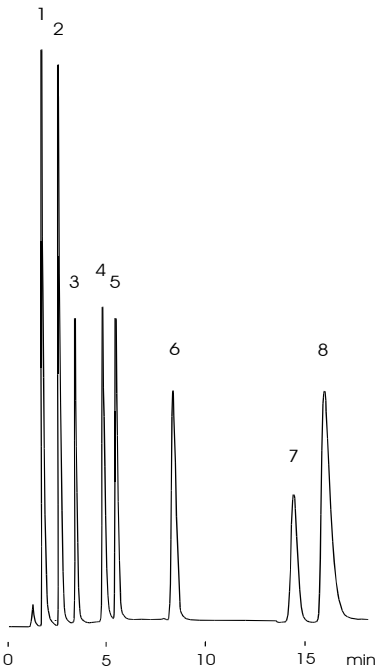


Fig. 1 Nucleo bases and nucleosides

Packing:	UltraSep ES PFP
Column:	150 x 3mm
Detection:	254 nm
Eluent:	15mM Phosphoric acid, pH 3.0
Flow rate:	0.6ml/min
Temperature:	30 °C

1 Cytosine; 2 Cytidine; 3 Uridine; 4 Guanine;
5 Thymine; 6 Guanosine; 7 Thymidine; 8 Adenosine
In comparison to RP 18 all components are
separated also on the short column without
problems, vgl. B 10.

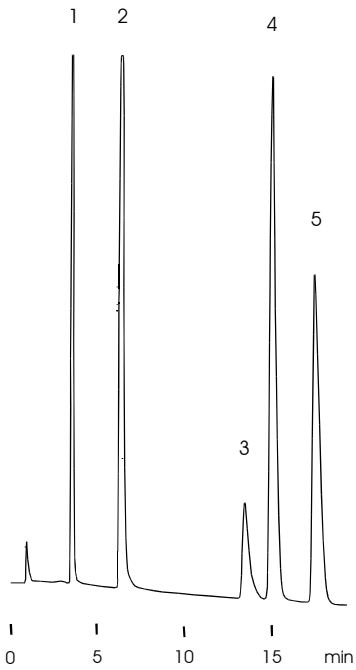


Fig. 2 Strong Bases

Packing:	UltraSep ES PFP
Column:	150 x 3mm
Detection:	215 nm
Eluent:	15mM Phosphor.ac./AN 7/3, pH 3.3
Flow rate:	0.6ml/min
Temperature:	40 °C

1 Mexiletine; 2 Clozapine,; 3 Perazine dimalonate;
4 Protriptyline; 5 Flecainide;

Pay attention to the excellent shape of the perazine peak. The order of 4 and 5 is reversed in comparison to normal RP packings., vgl. B 3.

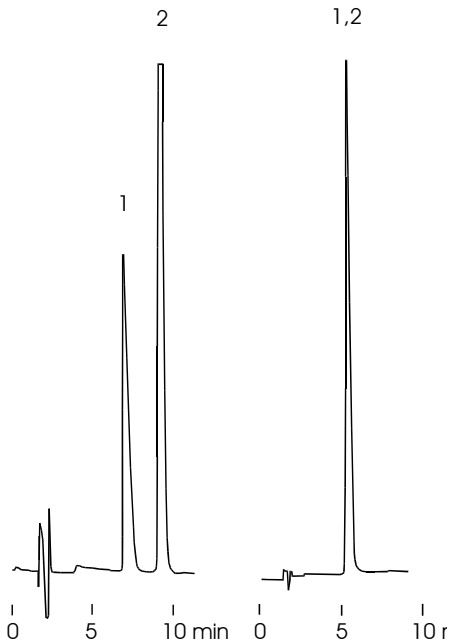


Fig. 3 Ephedrine/Ephedrone

Packing left:	UltraSep ES PFP
Packing right:	UltraSep ES PHARM RP18
Column:	150 x 3mm
Detection:	254nm
Eluent:	AN/water 15/85 + 0.5% TFA
Flow rate:	0.45ml/min
Temperature:	30 °C

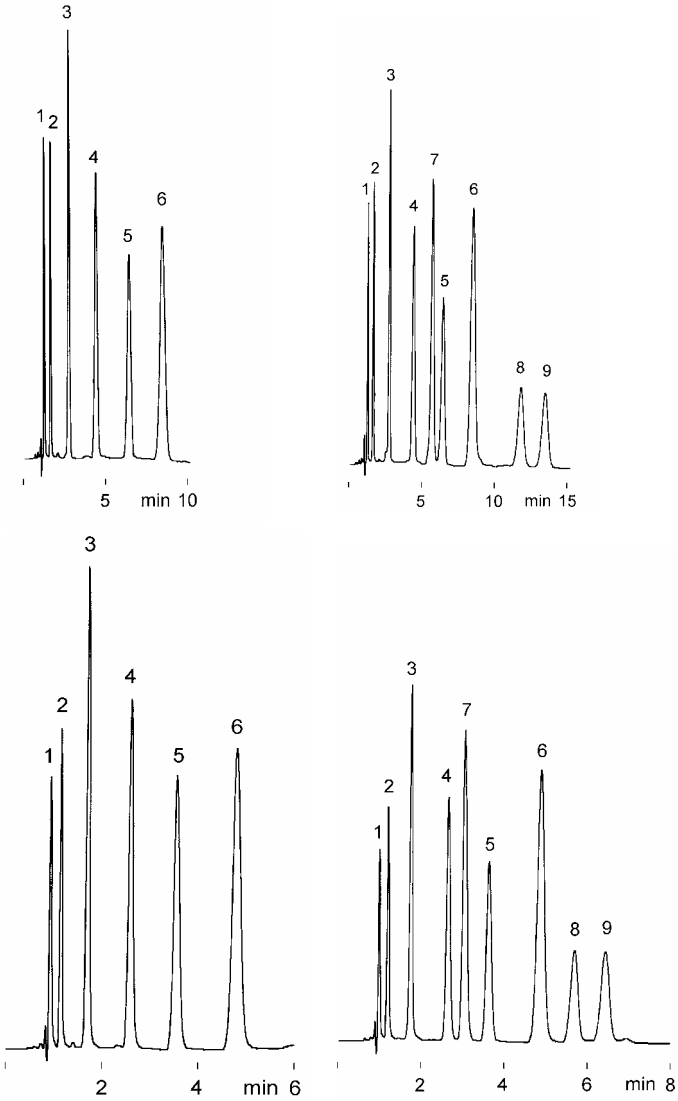
1 Ephedrine; 2 Ephedrone (CAT);

Prices please see O - 6

UltraSep ES CA and **UltraSep ES CAQC** (Catecholamines)

The special packings UltraSep ES CA and UltraSep ES CAQC were developed for the analysis of **catecholamines** (including their **metabolic products**) and **serotonine** in samples of urine. Columns for normal as well as for quick analyses (QC: Quick Columns) are offered.

Additionally, we offer columns for clinical-chemical routine analyses, compatible with the sets of other suppliers.



UltraSep ES CA

Packing:	UltraSep ES CA
Column:	125 x 3 mm
Detection:	UV, 280 nm
Eluent:	Eluent CA
Flow rate:	1.0 ml/min
Temperature:	25 °C

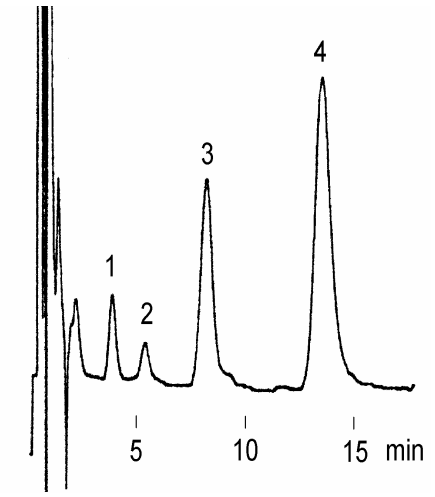
1 Vanillicmandelic acid (VMA)
2 Iso-VMA
3 3,4-Dihydroxyphenylacetic acid (DOPAC)
4 5-Hydroxyindolylacetic acid(5-HIAA)
5 Homovanillic acid(HVA)
6 5-Hydroxyindol-2-carbonic acid (5-HICA)
7 3-Hydroxytyramine (DOPAMIN)
8 3-Methoxytyramine (3-MT)
9 5-Hydroxytryptamine (SEROTONINE)

UltraSep ES CAQC

Packing:	UltraSep ES CAQC
Column:	125 x 3 mm
Detection:	UV, 280 nm
Eluent:	Eluens CA
Flow rate:	1.0 ml/min
Temperature:	25 °C

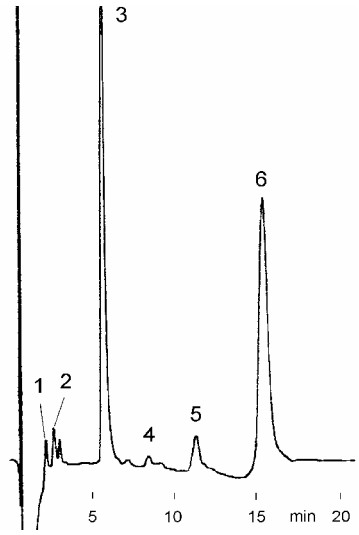
UltraSep ES CAAM and **UltraSep ES CAAC** (Catecholamines)

UltraSep ES CAAM is used for the **clinical-chemical routine analysis** of catecholamines, UltraSep ES CAAC to analyse the metabolic acids from catecholamines.



1 Noradrenaline
2 Adrenaline
3 3,4-Dihydroxybenzylamine (DHBA; Standard)
4 3-Hydroxytyramine (DOPAMINE)

Sample:	Urine of a patient (Charité Berlin)
Packing:	UltraSep ES CAAM
Column:	125 x 3 mm
Detection:	Electrochemical detector
Eluent:	Eluent CAAM
Flow rate:	0.75 ml/min
Temperature:	Room temperature



1 Vanillicmandelic acid (VMA)
2 Iso-VMA
3 3,4-Dihydroxyphenylacetic acid (DOPAC)
4 5-Hydroxyindolylacetic acid (5-HIAA)
5 Homovanillic acid (HVA)
6 5-Hydroxyindol-2-carbonic acid (5-HICA)

Sample:	Urine of a patient (Charité Berlin)
Packing:	UltraSep ES CAAC
Column:	125 x 3 mm
Detection:	Electrochemical detector
Eluent:	Eluent CAAC
Flow rate:	0.8 ml/min
Temperature:	Room temperature

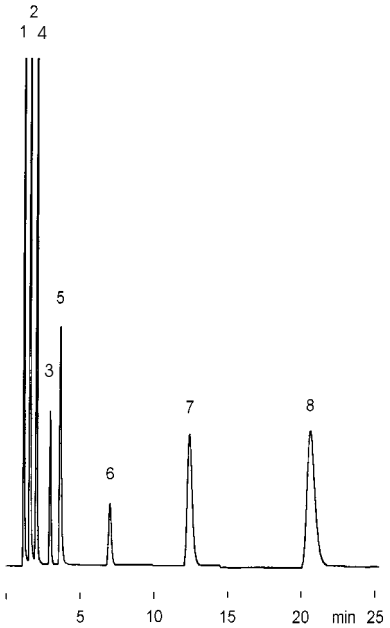
COLUMNS and **PRICES** (€/piece)

	Column size (mm)		
	125 x 4	125 x 3	125 x 2
UltraSep ES CA	335.00	335.00	335.00
UltraSep ES CAQC	335.00	335.00	335.00
UltraSep ES CAAM	335.00	335.00	335.00
UltraSep ES CAAC	335.00	335.00	335.00
Precolumn cartridges (mm)	10x4	10x3	10x2
VSK CA und CAAM	170.00	170.00	170.00
VSK CAQC	170.00	170.00	170.00
VSK CAAC	170.00	170.00	170.00
(Price per pack = 5 pieces)			
Eluent for CAAM [1])		45.00 €/l	
Eluent for CAAC [1])		45.00 €/l	

[1]) The composition of the eluent can be communicated to our customers on request.

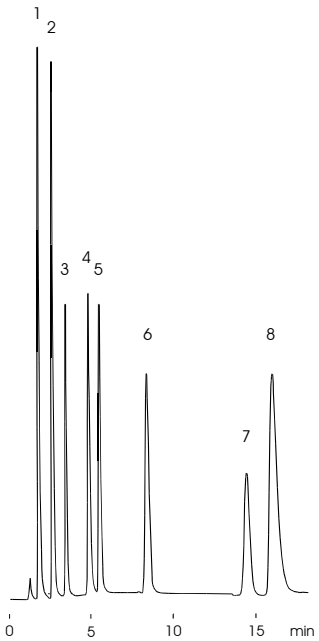
Analysis of Nucleo Acid Components

Mixtures of nucleobases, nucleosides, and nucleotides (e. g. hydrolysates of nucleic acids) can be separated by the high selective UltraSep ES PHARM RP18 packings as well as by UltraSep ES PFP. In the figures below the chromatograms of a mixture of nucleobases and nucleosides under isocratic conditions are presented. If there are more complicated hydrolysates, eluent gradients are used. In the same manner nucleotides can be separated using the ion pairing agent tetrabutylammonia hydrogensulfate. Nucleotides are analyzed effectively by anion exchangers (SAX) using a buffer gradient.



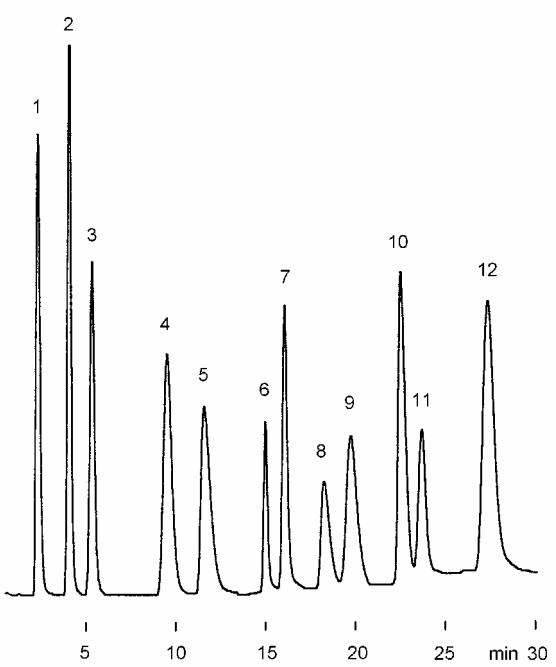
Packing:	UltraSep ES PHARM RP18E
Column:	250 x 3 mm
Detection:	UV, 254 nm
Eluent:	50 mM Phosphoric acid, pH 4.6 (NaOH)
Flow:	1.0 ml/min
Temperature:	35 °C

1 Cytosine
2 Cytidine
3 Uridine
4 Guanine
5 Thymine
6 Guanosine
7 Thymidine
8 Adenosine



Packing:	UltraSep ES PFP
Column:	150 x 3 mm
Detection:	254 nm
Eluent:	15 mM Phosphoric acid pH 3.0
Flow rate:	0.6 ml/min
Temperature:	30 °C

The following picture demonstrates a separation of Nucleosid-5'-mono-, -di- und -triphosphates at UltraSep SAX NN (s. S. I 3).



1	CMP	Cytidine-5'-monophosphate
2	UMP	Uridine-5'-monophosphate
3	GMP	Guanosine-5'-monophosphate
4	AMP	Adenosine-5'-monophosphate
5	CDP	Cytidine-5'-diphosphate
6	UDP	Uridine-5'-diphosphate
7	GDP	Guanosine-5'-diphosphate
8	CTP	Cytidine-5'-triphosphate
9	ADP	Adenosine-5'-diphosphate
10	UTP	Uridine-5'-triphosphate
11	GTP	Guanosine-5'-triphosphate
12	ATP	Adenosine-5'-triphosphate

Packing:	UltraSep ES SAX NN
Column:	60 x 3 mm
Detection:	UV, 254 nm
Flow rate:	0.8 ml/min
Temperature:	30 °C
Eluent A:	3 mM $NH_4H_2PO_4$, pH 4.4
Eluent B:	200 mM $NH_4H_2PO_4$, pH 4.4
Gradient:	Start 0% B
	13,0th min 7% B
	13,1th min 28% B
	20.2th min 29% B
	20.3th min 53% B
	28.0th min 85% B

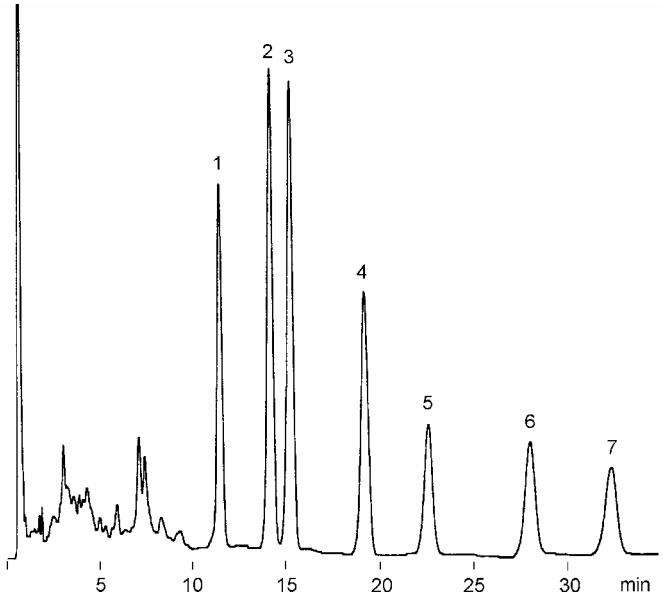
COLUMNS and **PRICES** (€/piece)

	Column size (mm)					
	250 x 3	250 x 2	125 x 3	125 x 2	60 x 3	60 x 2
UltraSep ES SAX NN	360.00	370.00	310.00	320.00	265.00	275.00
	Size (mm)					
	10 x 3	10 x 2	5 x 3	5 x 2		
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00	170.00		

Prices for PHARM Packings see pages **B-4**

Price for UltraSep ES PFP see page **O-6**

UltraSep ESD CHOL (Cholesterol Esters)



1 Cholesterol (Chol-OH)
2 Chol-Arachidonic acid
3 Chol-Linolenic acid
4 Chol-Linolic acid
5 Chol-Myristic acid
6 Chol-Oleic acid
7 Chol-Palmitic acid

Packing:	UltraSep ESD CHOL 3 µm
Column:	125 x 3 mm
Detection:	UV 206 nm
Eluent:	Acetonitrile/isopropanol (7/3)
Flow rate:	1 ml/min
Temperature:	25 °C

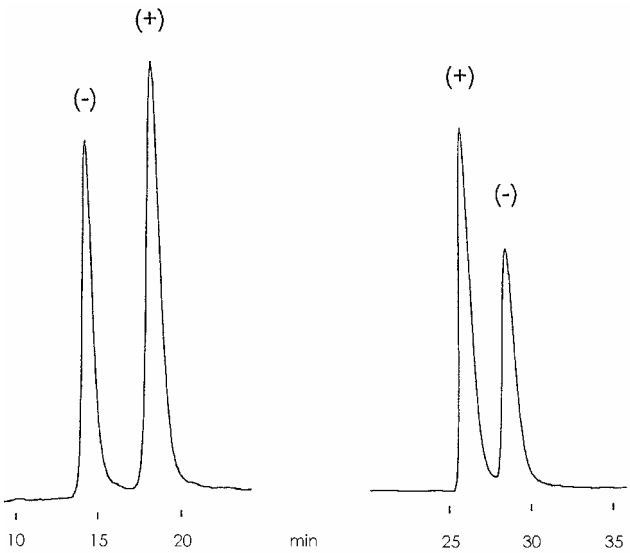
COLUMNS and **PRICES** (€/piece)

	Column size (mm)					
	250 x 4	250 x 3	250 x 2	125 x 4	125 x 3	125 x 2
UltraSep ESD CHOL	360.00	340.00	355.00	295.00	280.00	290.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2	5 x 2		
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00	170.00		

CHIRAL ANALYSIS

UltraSep ES CHIR P (Enantiomers)

In UltraSep ES CHIR P (P means Pirkle) 3,5-DNB-D-α-phenylglycine is coupled to the gel matrix via a spacer by an amide type bond (CHIRP, D-Phegly). The 3,5-dinitrobenzoyl (DNB) residue acts as a π-electron acceptor yielding good separation conditions for enantiomers with π-electrons. For some separations the analogously constructed DNB-L-leucine phase with a different selectivity is of advantage (CHIRP, L-Leu). This shows the peak reversal of the (+) and (-) forms of 2,2,2-trifluoro-1-(9-anthryl)ethanol on both packings.



Left:
Packing: UltraSep ES CHIR P,D-Phegly
Column: 125 x 2 mm
Eluent: n-Hexane/isopropanol/ acetonitrile (96/3/1)
Flow rate: 0.2 ml/min
Detection: UV 254 nm

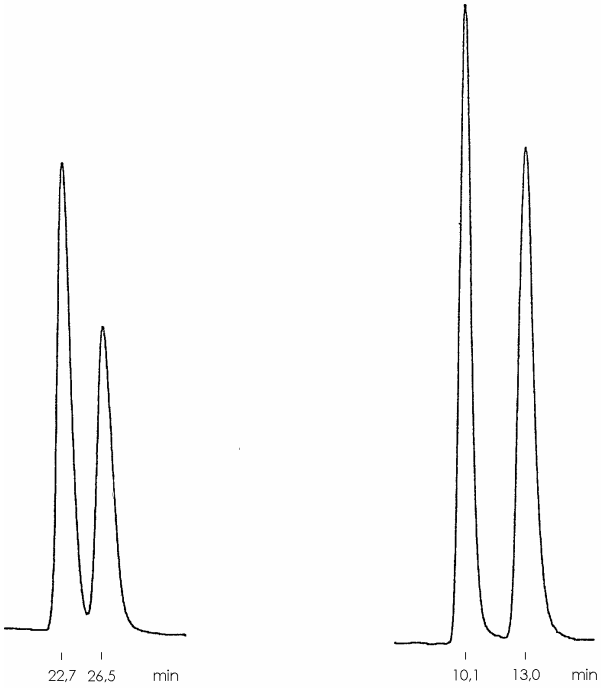
Right:
Packing: UltraSep ES CHIR P,L-Leu
Column: 250 x 2 mm
Eluent: n-Hexane/isopropanol/ acetonitrile (97/1/2)
Flow rate: 0.2 ml/min
Detection: UV 254 nm

COLUMNS and **PRICES** (€/piece)

	Column size (mm)			
	250 x 3	250 x 2	125 x 3	125 x 2
UltraSep ES CHIR P,D-Phegly			410.00	390.00
UltraSep ES CHIR P,L-Leu	530.00	465.00		

UltraSep ES CHIR PMA (Enantiomers)

This packing has a layer of polymeric methacrylic acid (PMA) on silica. It is characterized by hydrophobic properties. The figure below shows the separation of 2,2,2-trifluoro-1-(9-anthryl)ethanol, which is often used as "chiral probe" (left). As a practical example the separation of the optical isomers of the diuretic Cyclopenthiazid (Navidrex) is also demonstrated (right).



Left:
Packing: UltraSep ES CHIR PMA
Column: 125 x 2 mm
Eluent: n-Hexane/acetonitrile (99.7/0.3)
Flow rate: 0.8 ml/min
Detection: UV 254 nm

Right:
Packing: UltraSep ES CHIR PMA
Column: 125 x 2 mm
Eluent: n-Hexane/isopropanol (80/20) + 7 Vol.% acetonitrile
Flow rate: 0.4 ml/min
Detection: UV 254 nm

COLUMNS and **PRICES** (€/piece)

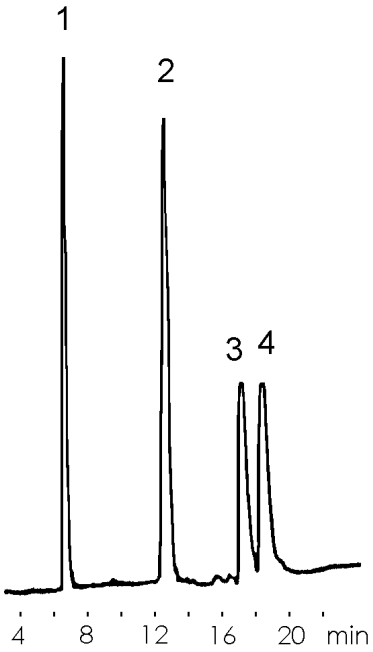
	Column size (mm)			
	250 x 3	250 x 2	125 x 3	125 x 2
UltraSep ES CHIR PMA	620.00	590.00	560.00	495.00

UltraSep ES RP Hypro Cu (Amino Acid Enantiomers)

UltraSep ES RP Hypro Cu was developed especially for the chiral separation of amino acids and derivatized amino acids.

Underivatized amino acids can be separated into their enantiomers by ligand exchange chromatography. The special problem was besides, to determine the N-acetyl compounds. All commercial ligand exchangers as far as checked were not able to do this.

For this reason SEPSERV developed a permanently loaded ligand exchange carrier on basis of RP . The combination of two chromatographical effects made it possible to achieve the wanted separation.



1 L-Methionine
2 D-Methionine
3 L-Acetylmethionine
4 D-Acetylmethionine

Packing:	UltraSep ES RP Hypro Cu
Column:	250 x 4 mm
Eluent:	1mM $CuSO_4$ solution in water
Flow rate:	1 ml/min
Detection:	UV 215 nm

COLUMNS and **PRICES** (€/piece)

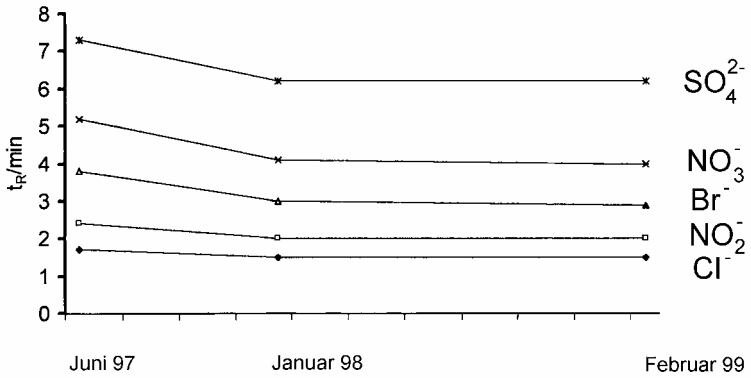
	Column size (mm)		
	250 x 4	250 x 3	250 x 2
UltraSep ES RP Hypro Cu	345.00	325.00	325.00
	Size (mm)		
	10 x 4	10 x 3	10 x 2
Precolumn cartridges (Price per pack = 5 pieces)	210.00	195.00	195.00

ION EXCHANGERS

Anion Exchangers

UltraSep ES SAX W (Anorganic ions according to DIN)

These anion exchangers are so called „permanently loaded“ ion exchangers. They exhibit the excellent performance of silicas and the good stability of RP-phases, which leads to low costs. They can be used for the determination of Anorganic ions according to DIN 38405.

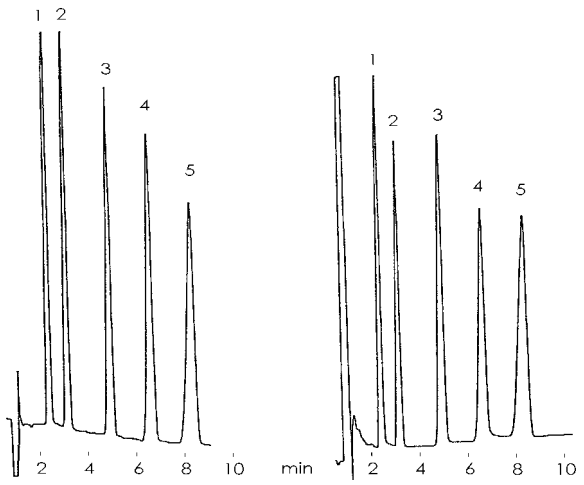


Stability of UltraSep ES SAXW with phthalic acid/TRIS buffer (1,25/2,5 mM) at pH 6.4

Permanently loaded ion exchangers are not only useful to separate inorganic ions (see below) but also to separate natural products with aqueous buffers (nucleotides, B 11). They cannot be used with organic-aqueous eluents.

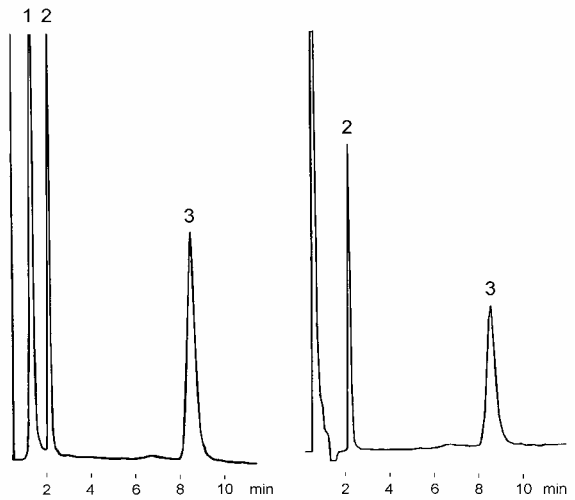
Our permanently loaded ion exchangers are easily to regenerate. For this, we offer a special solution or the regeneration in our firm.

If there is a need to chromatograph aqueous solutions highly contaminated with organic material, we recommend to use additionally a RP precolumn (length 33 mm) in front of the precolumn cartridge.



1 Chloride
2 Nitrite
3 Bromide
4 Nitrate
5 Sulfate

Packing:	UltraSep ES SAX W
Column:	125 x 3 mm
VSK	10 x 3 mm
Detection left:	UV 280
Detection right:	Conductivity Detectors in line
Eluent:	1.25 mM Phthalic acid 2.50 mM TRIS pH 6.5
Flow rate:	1.5 ml/min
Temperature:	30 °C

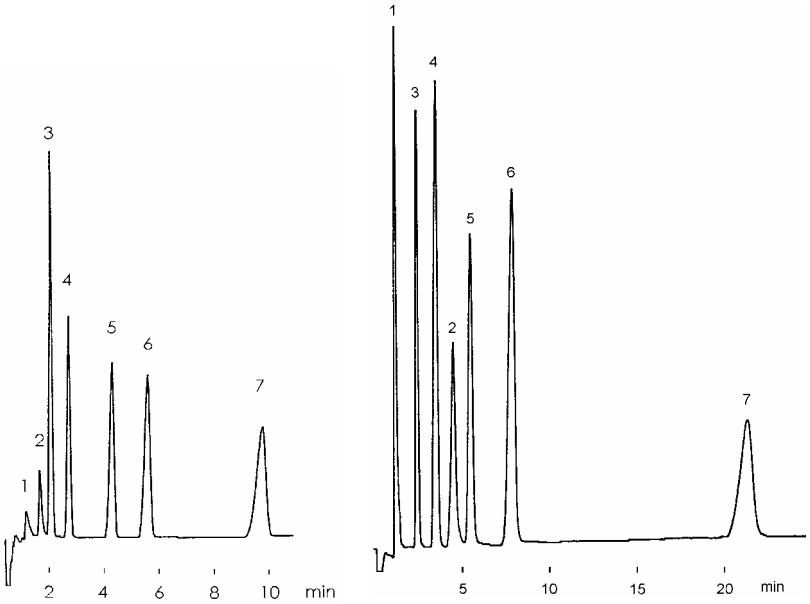


1 Carbonate
2 Chloride
3 Sulfate

Sample:	Tap water from Berlin
Packing:	UltraSep ES SAX W
Column:	125 x 3 mm with precolumn cartridge 10 x 3 mm
Detection:	UV 280 nm (left) conductivity (right) detectors in line
Eluent:	1.25 mM Phthalic acid 2.5 mM TRIS, pH 6.5
Flow rate:	1.5 ml/min
Temperature:	30 °C

Minimum detectability of 2 and 3 ≤ 1 mg/l

By lowering of the strength of the eluent, phosphate appears between nitrite and bromide without the necessity to increase the pH noticeably. In this way, there are good conditions for the conductivity detection too. Furthermore, fluoride is well separated from the injection peak.



1 Fluoride
2 Phosphate
3 Chloride
4 Nitrite
5 Bromide
6 Nitrate
7 Sulfate

Packing:	UltraSep ES SAX W
Column:	125 x 3 mm
Precolumn cartridge:	10 x 3 mm
Detection:	UV 280 nm
Eluent left:	1,25 mM Phthalsäure with TRIS to pH 5.8
Eluent right:	0.3 mM Phthalic acid with TRIS to pH 6.6
Flow rate:	1.5 ml/min
Temperature:	30 °C

For the same packing 50 mM sulfuric acid can be used as eluent. This is advantageous in the case of UV-active anions.

Columns and **Prices** (in **€**/ peace)	Separation Columns		Precolumns	Precolumn cartridges (Price per pack=5 pieces)	
	250x4mm 200x4mm	125 x 3mm	33 x 3mm	10 x 4mm	10x3mm
UltraSep ES SAX W		270.00			170.00
UltraSep ES SAX W Guard Column RP18 *)			125.00		
UltraSep ES SAX QCM **)	290.00	270.00		170.00	
Activator for ES SAX W (1ml)			12.00		
Regeneration solution (30 ml) for regeneration by customer			33.00		

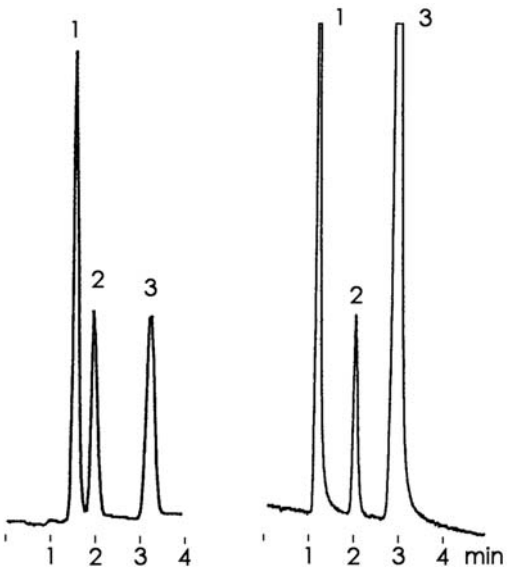
*) Recommended as precolumn for samples with high content of organic substances.
**) With chemical bonded quaternary exchanger groups.

UltraSep ES SAX NN (Rapid determination of Nitrite, Nitrate and Bromide in drinking water and food)

By this exchanger concentrations of nitrite and nitrate in drinking water and foodstuffs can be determined **considerably faster** and **more sensitive** than with customary methods. Nitrate, which can be determined by the photometric method only relatively insensitively, is determined by chromatography at 220 nm without previous enrichment to 0.1 mg/l. Bromide is determined at 210 nm with high sensitivity.

Iodide can be determined also directly at 225 nm. For a quick determination of iodide you can use a column of UltraSep ES SAX NN with a length of 33 mm.

The selectivity and rapidity of the determination (phosphoric acid as eluent) can be changed by the pH value (at best pH 3 – 5). Furthermore, the velocity of elution can also be varied by the concentration of phosphoric acid (at best 10 – 30 mM; see the following chromatograms).



1 Nitrite
2 Bromide
3 Nitrate

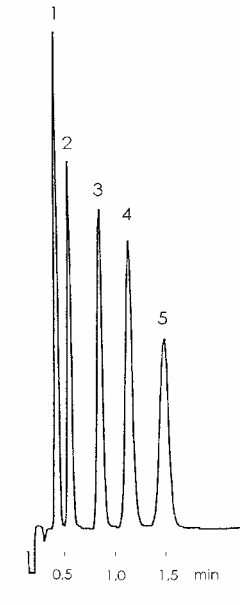
Left:
Packing: UltraSep ES SAX NN
Column: 60 x 3 mm
+ 10 x 3 mm Precolumn cartridge
Detection: UV 220 nm
Eluent: 10 mM Phosphoric acid pH 5 (with NaOH)
Flow rate: 1.0 ml/min
Temperature: Room temperature

Right:
Packing: UltraSep ES SAX NN
Column: 60 x 3 mm
+ 10 x 3 mm Precolumn cartridge
Detection: UV 220 nm
Eluent: 30 mM Phosphoric acid pH 3 (with NaOH)
Flow rate: 1.0 ml/min
Temperature: Room temperature

COLUMNS and **PRICES** (€/piece)

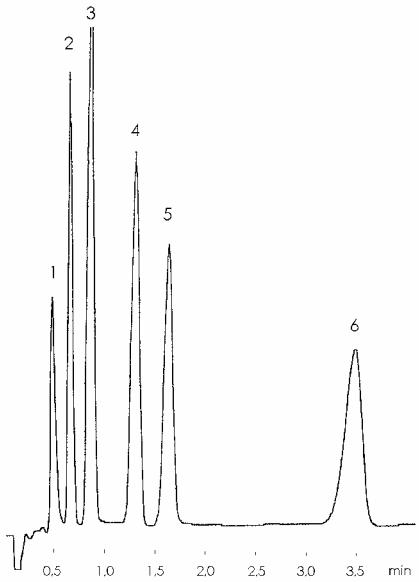
	Column size (mm)	
	60 x 3	33 x 3
UltraSep ES SAX NN	265.00	245.00
	Size (mm)	
	10 x 3	5 x 3
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00

UltraSep ES SAX W Quick Columns (Anorganic Anions)



1 Chloride
2 Nitrite
3 Bromide
4 Nitrate
5 Sulfate

Packing:	UltraSep ES SAX W QC
Column:	33 x 3 mm (no special demands on devices, no microcell)
Detection:	UV 280 nm
Eluent:	1.25 mM Phthalic acid 2.50 mM TRIS, to pH 6.5
Flow rate:	2 ml/min
Temperature:	30 °C



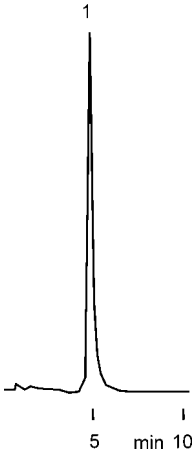
1 Phosphate
2 Chloride
3 Nitrite
4 Bromide
5 Nitrate
6 Sulfate

Packing:	UltraSep ES SAX W QC
Column:	60 x 3 mm
Detection:	UV 280 nm
Eluent:	1.25 mM Phthalic acid approx. 2 mM TRIS, to pH 5.6
Flow rate:	2 ml/min
Temperature:	30 °C

COLUMNS and **PRICES** (€/piece)

	Column size (mm)	
	60 x 3	33 x 3
UltraSep ES SAX W QC	255.00	250.00
	Size (mm)	
	5 x 3	10 x 3
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00

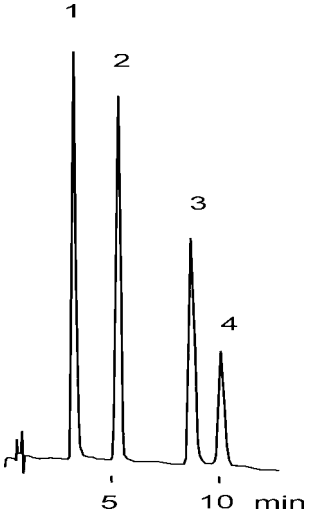
UltraSep ES SAX W33 (Ascorbic Acid)



1 Ascorbic acid

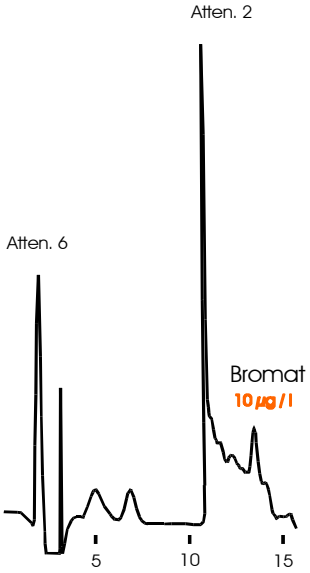
Packing:	UltraSep ES SAX W 33
Column:	125 x 3 mm
Detection:	UV 254 nm
Eluent:	10 mM Ammonium acetate pH 3.5
Flow rate:	0.6 ml/min
Temperature:	Room temperature

UltraSep ES RP NN (Nitrite, Bromate, Bromide, Nitrate)



1 Nitrite
2 Bromate
3 Bromide
4 Nitrate

Peak 1 corresponds to 2,0 mg/ l
(injection volume: 10 µl)
Peak 4 corresponds to 0.5 mg / l
(injection volume: 10 µl)



Packing:	UltraSep ES RP NN
Column:	150 x 3 mm
Detection:	UV 210 nm
Eluent:	1 mM Hexadecyltrimethylammonium-hydrogensulfate/acetonitrile (3/1)
Flow rate:	0.8 ml/min
Temperature:	30 °C

Bromate in Tap Water

Flow rate:	0.4 ml/min
Injection volume:	500 µl
Other conditions:	As described left

COLUMNS and **PRICES** (€/piece)

	Column size (mm)	
	125 x 3	150x3
UltraSep ES SAX W33	295.00	
UltraSep ES RP NN	240.00	240.00
	Size (mm)	
	10 x 3	
Precolumn cartridges (Price per pack = 5 pieces)	170.00	

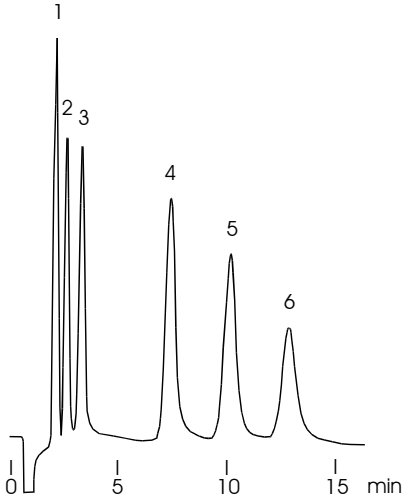
Cation Exchanger

All the cation exchangers offered possess chemical fixed exchanger groups and indicate a very good stability within acid media. Weak cation exchangers bear carboxylic groups, strong exchangers sulfonic groups, usually connected with phenyl residues. The letter H characterizes a special high sulfonated phenyl carrier with an exchange capacity of 0.6 mval/g. The letters PM indicate an ES gel, coated with a polymer layer, with aliphatically connected sulfo groups..

Weak cation exchangers WCX

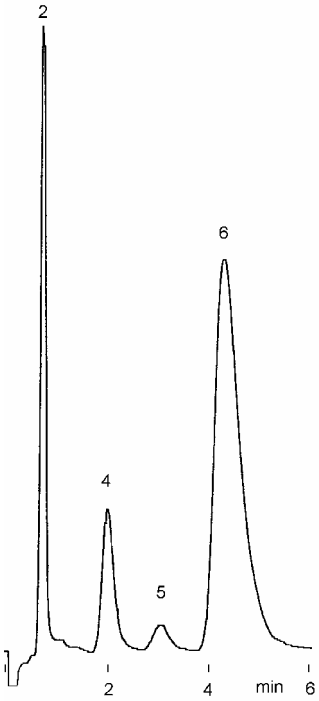
UltraSep ES WCX44 (Alkali, Alkaline Earth Ions, Ammonia)

With this packing alkali and alkaline earth ions can be separated in the presence of ammonia.



1 Li^+
2 Na^+
3 $NH4^+$
4 Mg^{++}
5 K^+
6 Ca^{++}

Packing: UltraSep ES WCX44
Column: 125 x 3 mm
Detection: Conductivity
Flow rate: 0.75 ml/min
Temperature: 30 °C
Eluent: 0.5 mM Citric acid
0.25 mM Oxalic acid
3 mM 18-Crown-6

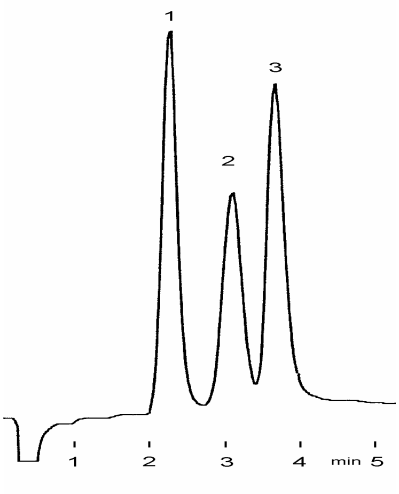


Quick Analysis of Drinking and Industrial Water

2 Na^+
4 Mg^{++}
5 K^+
6 Ca^{++}

Sample: **Tap water from Berlin**
Packing: UltraSep ES WCX44
Column: 60 x 3 mm
Detection: Conductivity
Eluent: 1 mM Citric acid
0.5 mM Oxalic acid
15 mM 18-Crown-6
Flow rate: 1.5 ml/min
Temperature: 50 °C

UltraSep ES WCX56 (Hydroxylamine, Ammonia)

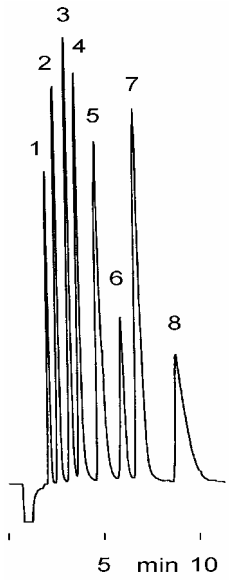


1 Na^+
2 $NH3^+OH$
3 $NH4^+$

Packing: UltraSep ES WCX56
Column: 125 x 3 mm
Detection: Conductivity
Eluent: 1 mM Citric acid
0.5 mM Oxalic acid
15 mM 18-Crown-6
Flow rate: 1.5 ml/min
Temperature: 30 °C

UltraSep ES WCXC (Alkali, Alkaline Earth Ions, Ammonia)

In contrast to the exchangers WCX44 and WCX56 in WCXC the component 18-crown-6 is already chemically fixed at the silica gel surface together with the exchanger groups (carboxylic groups). Therefore one can omit its addition to the mobile phase.



1 Li^+
2 Na^+
3 $NH4^+$
4 Cs^+
5 Rb^+
6 Mg^{++}
7 Ca^{++}
8 K^+

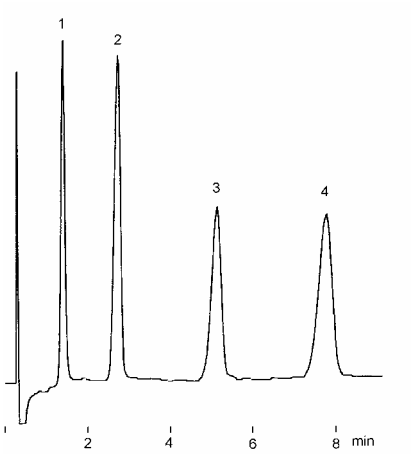
Packing: UltraSep ES WCXC
Column: 250 x 3 mm
Detection: Conductivity
Eluent: 4 mM Oxalic acid pH 2.4
Flow rate: 1.0 ml/min
Temperature: 30 °C

COLUMNS and **PRICES** (€/piece)

	Column size (mm)			
	125 x 3	125 x 2	60 x 3	60 x 2
UltraSep ES WCX44	590.00	590.00	435.00	435.00
UltraSep ES WCX56	590.00	590.00	435.00	435.00
UltraSep ES WCXC	590.00	590.00	435.00	435.00
	Size (mm)			
	10 x 3	10 x 2		
Precolumn cartridges (Price per pack = 5 pieces)	195.00	195.00		

Strong cation exchangers SCX

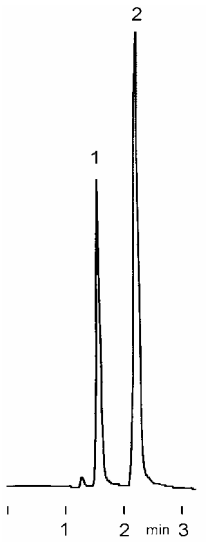
UltraSep ES SCX75 (Alkaline Earth Ions)



1 Mg^{++}
2 Ca^{++}
3 Sr^{++}
4 Ba^{++}

Packing: UltraSep ES SCX75
Column: 125 x 3 mm
Detection: Conductivity
Eluent: 5 mM Oxalic acid
5 mM Ethylene diamine
Flow rate: 1.5 ml/min
Temperature: 30 °C

UltraSep ES SCXPM



1 Maleic acid
2 Fumaric acid

Packing: UltraSep ES SCXPM
Column: 200 x 3 mm
Detection: UV 210 nm
Eluent: 5 mN H_2SO_4
Flow rate: 0.45 ml/min
Temperature: 30 °C

COLUMNS and **PRICES** (€/piece)

	Column size (mm)					
	250 x 4	250 x 3	250 x 2	125 x 4	125 x 3	125 x 2
UltraSep ES SCXPM	Prices	300.00	315.00	300.00	285.00	305.00
UltraSep ES SCX75	on	240.00	255.00	240.00	220.00	240.00
UltraSep ES SCXH	Request	250.00	265.00	250.00	235.00	250.00
	Size (mm)					
	10 x 4	10 x 3	10 x 2			
Precolumn cartridges (Price per pack = 5 pieces)	195.00	195.00	195.00			

OTHER SPECIAL COLUMNS

Special columns are a central concern in our company. With an appropriate column you can solve nearly all separation tasks, not laboriously by high plate numbers, but simply by tailor made selectivities. Tell us your problem!

By the way, the following packings are only available from SEPSERV. They were mostly developed due to orders of customers. For reasons of discretion it is not possible in every case to name the separation problems worked on.

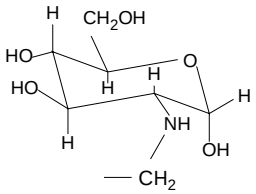
UltraSep ES POLYOL, optically active as

UltraSep ES AMINOPOLYOL H — CH_2 — NH — CH_2 — $(CHOH)_4$ — CH_2OH

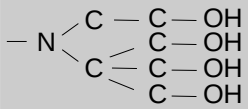
(Aminodesoxyhexitol)

UltraSep ES AMINOPOLYOL G

(Aminodesoxy-D-glucose)



UltraSep ES AMINOTETRAOL



Alcoholic hydroxyls and amino groups on the surface of this packing are responsible for a blocking of residual silanols. This means, that polar compounds can be excellently separated by normal phase chromatography. Example: Stereoisomeric organic N-oxides. AMINOTETRAOL is a very versatile phase. In the pharmaceutical industry it is advantageously used also with aqueous and aqueous-organic eluents. One can use the phase to separate sugars and more. Furthermore, the packing works as a weak ion exchanger and separates with weak eluents, e. g., organic carbon acids at pH 3.

UltraSep ES DTA

(Diethylenetriamine) – CH_2 – NH – CH_2 – CH_2 – NH – CH_2 – CH_2 – NH_2

This packing is designated for special applications with amino packings. To this group belongs also UltraSep ES TETRAMINOOL

UltraSep ES TETRAMINOOL (TTA)

(Triethylenetetramine) – CH_2 – NH – $(CH_2$ – CH_2 – NH$)_2$ – CH_2 – CH_2 – NH_2

This packing has in comparison with DTA a doubled spacer chain and three ethylene residues connected by nitrogen atoms. Beside the primary amino group at the end of the chain there exists an alcoholic hydroxyl group at the spacer.

UltraSep ES RP8F

(Fluoroalkyl) – $(CF_2)_7$ – CF_3

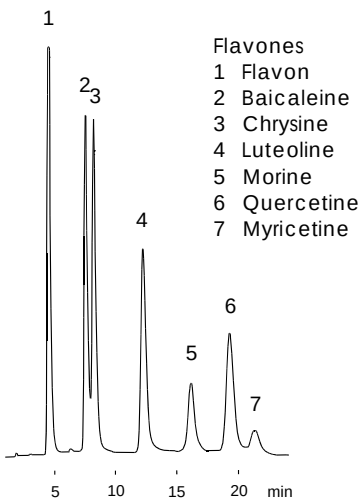
This packing is a fluorooctylsilica, where the H-atoms of each C-atom are substituted by 17 fluorine atoms. This packing is an alternative RP phase with relatively rigid strong hydrophobic chains for special applications.

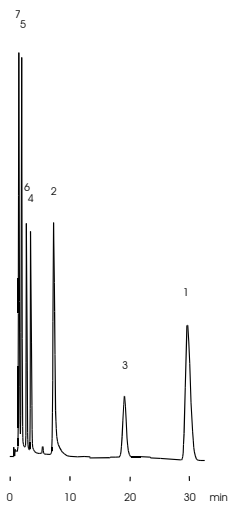
UltraSep ES PEO

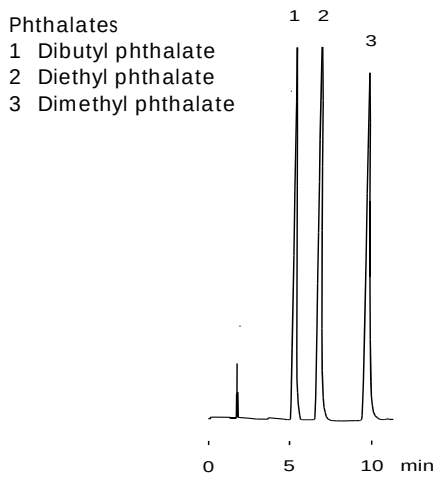
(Polyethyleneglycolsilica)

$$-(CH_2-CH_2-O)_{6\text{-}9}-CH_2-CH_2-O-CH_3$$

UltraSep ES PEO is a packing on the basis of polyethyleneoxide chains. Within the chain C2 segments alternate with oxygen atoms. In total there are 26 atoms within the chain, 18 of them are carbon atoms as it is the case in standard reversed phases. Their properties are levelled by the oxygen atoms and combined with interactions due to hydrogen bridges (OH....O; NH....O). Therefore the PEO packing can be used both for reversed phase chromatography and for normal phase chromatography (see below, left and right chromatogram, respectively). With adequate compounds it exhibits specific selectivity effects as well as shorter retention times in comparison to RP18 materials (see comparative chromatogram in the middle).







	Left	Middle
Packing:	UltraSep ES PEO	UltraSep ES RP18
Column:	150 x 3 mm	150 x 3 mm
Detection:	UV 254 nm	UV 254 nm
Eluent:	Methanol/water 4/6 + 0.5% TFA	Methanol/water 1/1 + 0.5% TFA
Flow rate:	0.35 ml/min	0.8ml/min
Temperature:	30°C	30°C

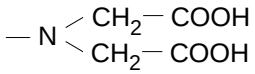
	Right
Packing:	UltraSep ES PEO
Column:	150 x 3 mm
Detection:	UV 254 nm
Eluent:	n-Heptane + 0.5% i-propanol
Flow rate:	0.3 ml/min
Temperature:	Room temperature

UltraSep ESD300 C30

(RP-packing) $-(CH_2)_{29}-CH_3$

For the long C30 chains a basis material was chosen with an average pore diameter of 300 Å.

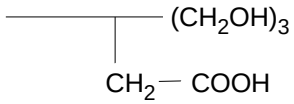
UltraSep ES ADC



(Aminodicarboxy)

As active group aminodiacetic acid was bound over a spacer with 6 C atoms.

UltraSep ES TC



(Triolcarboxy)

In this packing a carboxylic group was combined with a hydroxymethyl cluster having the basic ligand structure shown in the formula.

UltraSep ES C12 TA

(Triamino)

This packing is strongly polar and possesses branched aliphatic chains containing two terminal primary amino groups and one secondary amino group.

UltraSep ES RP18 M500 *)

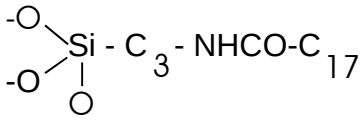
This packing is a lowly charged methyloctadecylsilica gel with a specific selectivity

SHIELD – CARRIER (EMBEDDED GROUPS)

RP packings with polar selectivity, suitable

- for purely aqueous media ("Aq") without phase collaps
- for the separation of highly polar compounds

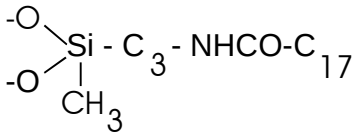
UltraSep ES AMID RP18A *)



[3-(Octadecanoylamino)propylsilyl-Silica]

The packing carries an amide group between spacer and RP chain (so called embedded group). **Specific area 280 m²/g.**

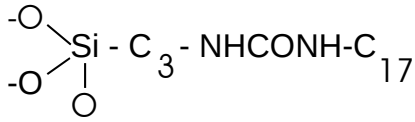
UltraSep ES AMID RP18AM



[3-(Octadecanoylamino)propylmethylsilyl-Silica]

The packing possesses additionally a **methyl group** at the silyl residue. **Specific area 420 m²/g.**

UltraSep ES AMID H RP18PA *)



3-(Heptadecylureido)-propylsilyl-Silica

The packing carries a urea group instead of the amide group.

*) See GIT **46** (2002), number 1; please ask for a reprint

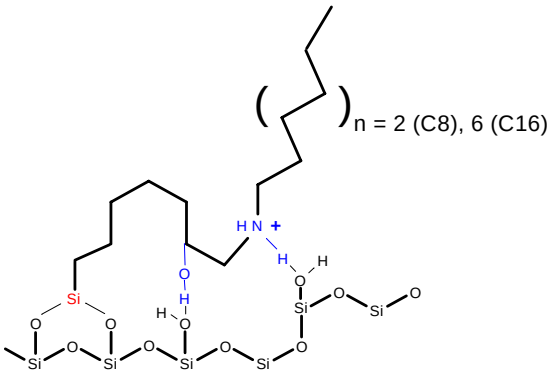
UltraSep ES CHAIR *)

UltraSep ES CHAIR is a special phase obtained by chemical binding of a sufficiently long non polar chain to the silica. The chain bears one or two polar substituents at the first third or at the middle part.

The groups show a pronounced interaction with silanols. Therefore, depending on the chromatographic conditions for a unique chain folding, a chair like configuration can be expected (see figure).

It is well known, that normal phases with polar groups show RP-behaviour too. With chair phases, that dual behaviour was intentional, so to speak, typified. In this way we can synthesize tailor-made selectivities, changing the length of the non polar chain or the nature and/or number (1 or 2) of the polar groups.

UltraSep ES CHAIR is often used, e. g. in the pharmaceutical industry and in biochemical institutions. Example: Cephalosporines.



Structure of UltraSep ES CHAIR

(on principle)

UltraSep ES UREIDO

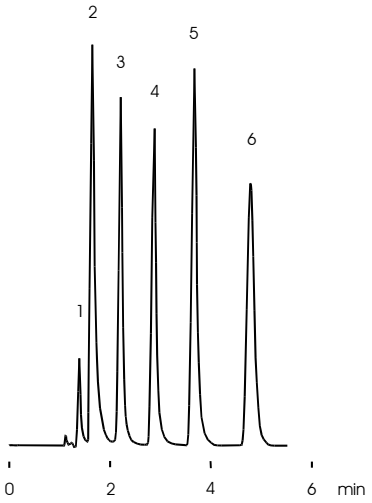
(Ureidopropylsilyl-silica)

Normal phase with a urea group at the end of the propyl spacer.

UltraSep ES C10 ω-COOH

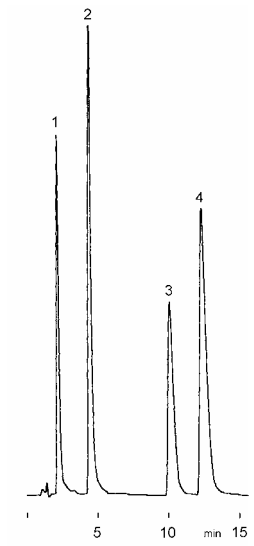
This is a medium polar packing with an unbranched aliphatic carbon chain and two carboxylic groups, one is terminal. It shows typical RP behaviour.

*) See GIT **46** (2002), number 1; please ask for a reprint



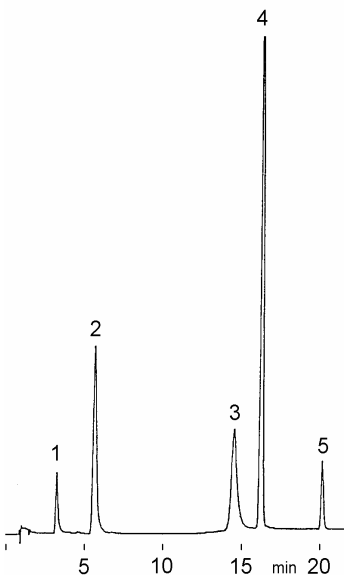
1 Uracil
2 Pyridine
3 Phenol
4 N,N-Dimethylaniline
5 Toluene
6 4-Butylbenzoic acid

Packing: UltraSep ES AMID H RP18PA
Column: 150 x 3 mm
Detection: 254 nm
Eluent: 50 mM Phosphoric acid pH 3.2/ acetonitrile (35/65)
Flow rate: 0.42 ml/min
Temperature: Room temperature



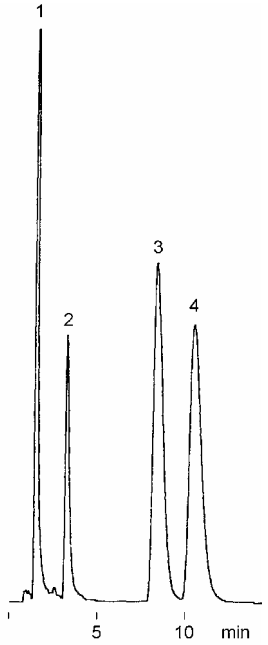
1 Clozapin
2 Perazine
3 Protriptyline
4 Amitriptyline

Packing: UltraSep ES AMID RP18A
Column: 125 x 2 mm
Detection: 254 nm
Eluent: 10 mM Trifluoroacetic acid in water/ acetonitrile (70/30); pH 2.2
Flow rate: 0.25 ml/min
Temperature: 36 °C



Nucleobases
1 Impurity from 4
2 6-Mercaptopurine
3 6-Thioguanine
4 6-Methylmercaptopurine
5 Impurity from 4

Packing: UltraSep ES AMID H RP18PA
Column: 150 x 3 mm
Detection: 254 nm
Eluent: A: 50 mM Phosphoric acid/ 1 mM octanesulfonate pH 2.5
B: Methanol
Gradient: Start 0 % B
5th min 0 % B
20th min 40 % B
Flow rate: 0.6 ml/min
Temperature: 40 °C



Packing: UltraSep ES CHAIR
Conditions and peak names as above right

COLUMNS and **PRICES** (€/piece)

UltraSep ES	Column size (mm)								
	250 x 4 200 x 4	250 x 3 200 x 3	250 x 2 200 x 2	150 x 4 125 x 4	150 x 3 125 x 3	150 x 2 125 x 2	100 x 4 60 x 4	100 x 3 60 x 3	100 x 2 60 x 2
AMINOPOLYOL H	265.00	265.00	270.00	255.00	255.00	260.00	245.00	245.00	250.00
AMINOPOLYOL G	265.00	265.00	270.00	255.00	255.00	260.00	245.00	245.00	250.00
AMINOTETRAOL	250.00	250.00	255.00	240.00	240.00	245.00	225.00	225.00	235.00
DTA	290.00	290.00	290.00	265.00	265.00	270.00	255.00	255.00	260.00
TETRAMINOOL (TTA)	290.00	290.00	290.00	265.00	265.00	270.00	255.00	255.00	260.00
RP8F	330.00	315.00	315.00	290.00	290.00	295.00	280.00	280.00	285.00
PEO	510.00	480.00	480.00	430.00	430.00	430.00	410.00	400.00	400.00
PFP	510.00	480.00	480.00	430.00	430.00	430.00	410.00	400.00	400.00
RP 30 and RP 30DM	395.00	395.00	395.00	365.00	365.00	365.00	355.00	355.00	355.00
D300 C30	480.00	480.00	480.00	445.00	445.00	445.00	435.00	435.00	435.00
ADC	265.00	265.00	270.00	255.00	255.00	260.00	245.00	245.00	250.00
TC	265.00	265.00	270.00	255.00	255.00	260.00	245.00	245.00	250.00
C12 TA	250.00	250.00	255.00	240.00	240.00	245.00	225.00	225.00	235.00
C10 ω-COOH	290.00	290.00	290.00	265.00	265.00	270.00	255.00	255.00	260.00
RP18 M500	270.00	265.00	265.00	240.00	235.00	235.00	210.00	210.00	215.00
AMID RP18A	340.00	335.00	335.00	305.00	300.00	300.00	285.00	285.00	290.00
AMID RP18AM	340.00	335.00	335.00	305.00	300.00	300.00	285.00	285.00	290.00
AMID H RP18P	270.00	265.00	265.00	240.00	235.00	235.00	210.00	210.00	215.00
AMID H RP18PA	270.00	265.00	265.00	240.00	235.00	235.00	210.00	210.00	215.00
CHAIR	240.00	240.00	245.00	235.00	225.00	235.00	215.00	215.00	220.00
UREIDO	210.00	210.00	210.00	210.00	200.00	200.00	200.00	200.00	200.00
Phenylhexyl	390.00	380.00	395.00	370.00	360.00	375.00	335.00	325.00	340.00
	Size (mm)								
	10 x 4	10 x 3	10 x 2						
Precolumn cartridges (Price per pack = 5 pieces)	170.00	170.00	170.00						

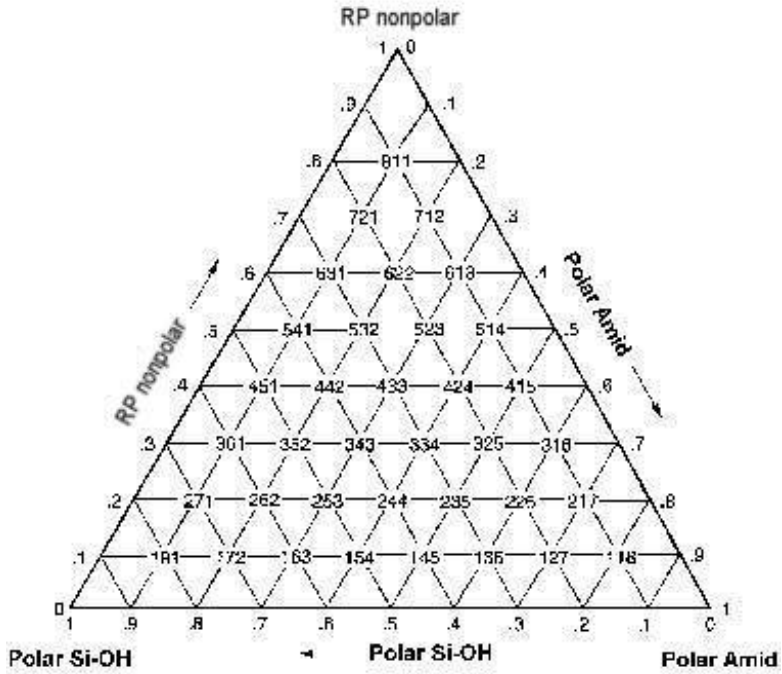
SELECTIVTY PACK TRIS

3 Selectivities with one set or with one column

Selectivity Pack TRIS and Column TRIS

The selectivity of the separation column is the most important optimization criterion of HPLC. Until now for the most used RP chromatography there didn't exist any column set with defined and sufficiently different selectivities enabling users to optimize their separations goal-directed.

This gap was closed by the selectivity pack **„**TRIS SEPSERV**“** (TRIS = TRI Selectivities), introduced by SEPSERV on the Analytica 2002. This set is made of 3 columns with different selectivities, each 150 x 3 mm. The price of the set is 500.00 € (**30 % advantage).**



The set contains:

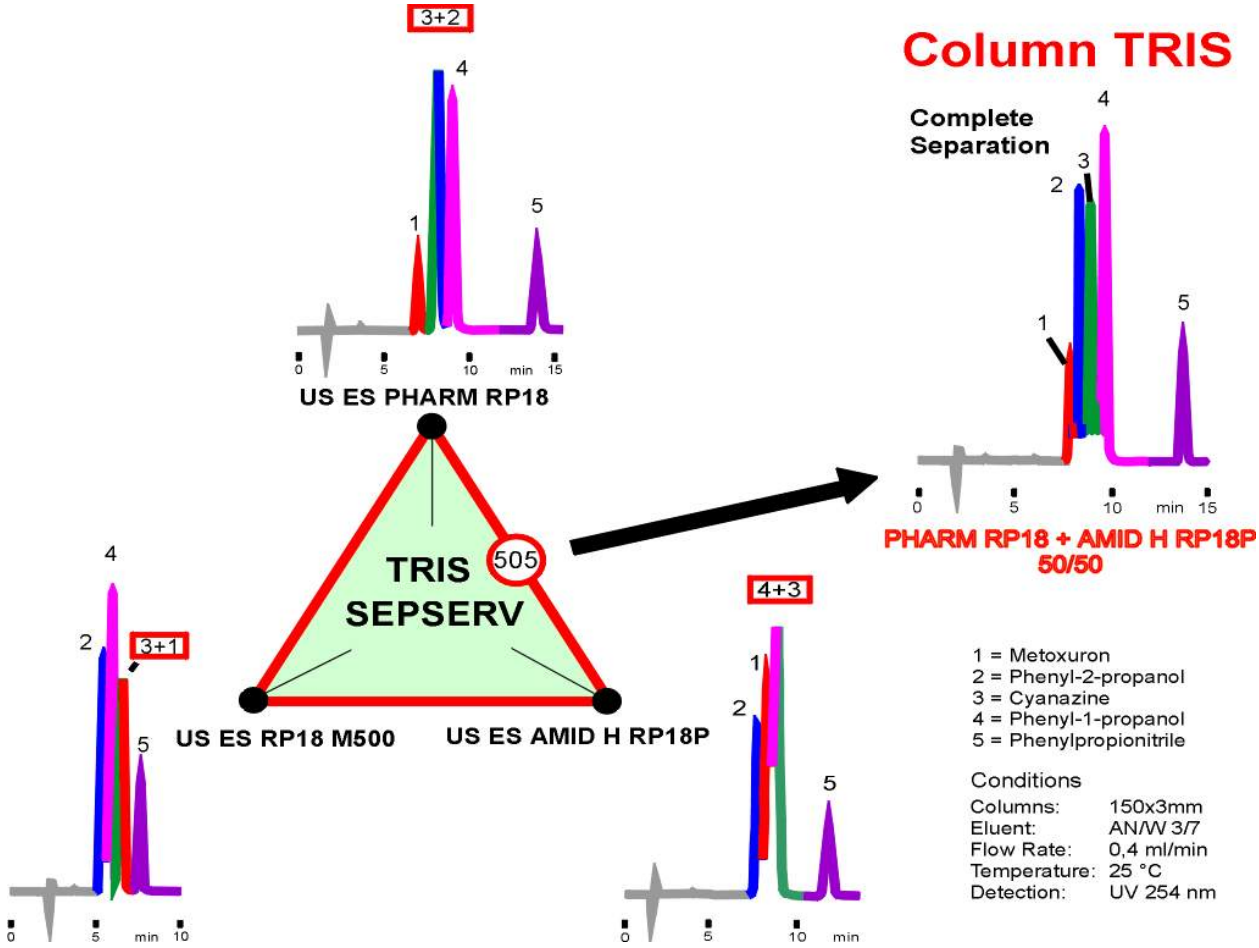
1) An UltraSep ES RP18 column of PHARM type, with high coverage (upper corner of the concentration/selectivity triangle),
2) a (special) column UltraSep ES RP18 M500 with a low coverage, exhibiting so called „Extended Polar (Matrix) Selectivity“, where silanol groups become effective (left corner of the triangle), and
3) a polar RP column of the type UltraSep ES AMID H RP18P with so called „Embedded Polar (Amid) Selectivity“ (right corner of the triangle), where embedded amid groups become effective and silanols are shielded [see **GIT 46 (2002) (1) 72**].

In addition to the set the column **„**Column TRIS**“** is offered for the first time by SEPSERV. The reason lies in the following: Sometimes the optimum separation cannot be obtained with any column of the set (corners of the triangle) because it needs a selectivity along a side of the triangle or coordinates within the triangle (for instance 352).

In this case, the optimum column can be easily calculated on the basis of the retention times of the 3 single columns isocratically obtained under identical conditions. By means of these retention times the customer can order a "tailor made" Column TRIS from SEPSERV.

Of course,optimizations of the packing and of the eluent can becombined with each other.

Application example see next page



COLUMNS and **PRICES** (€/piece)	Column size (mm)
	150 x 3
Selectivity Pack TRIS (3 Columns)	500.00
Column TRIS	250.00

Literature (Book Offer)

Eppert, Günter J.

HPLC TROUBLESHOOTING

Textbook and Training Manual for Chromatographers with 32 Exercises (Text in English and German)

(Available from book shops or from SEPSERV)

1. Edition
SEPSERV Separation Service, Berlin 2003
145 pages, 36 figures, 13 tables
Price: € 25.00
ISBN 3-00-010808-4

Preface

This book is a result of practical experience and is intended to assist in the reader's practical work. The material has been prepared from that of numerous training courses which I have conducted at SEPSERV and elsewhere. Many difficulties that clients have brought to our attention, as well as our own experience of more than twenty-five years with various issues and HPLC equipment have provided ample input.

My book focuses on the individual's "*quickly grasping a situation, then identifying and avoiding the problems*". Illustrations and tables that accompany the running text help provide the reader with a comprehensive overview of the material.

One ultimate goal after close study of this book is that the reader is capable of *actually judging separation columns* and not just immediately blaming the separation column every time the equipment fails or other trouble arises

Particular emphasis has been placed on the use of *narrow bore* columns with internal diameters of 3 and 2 mm, since there is no longer any reason compelling enough to make us have to get along without the advantages of using them in our daily work.

Successful troubleshooting requires a certain amount of prior knowledge in chromatography. Therefore, as an introduction and to broaden existing knowledge on the subject, I occasionally refer to my book, *"Flüssigchromatographie, HPLC – Theorie und Praxis"*.

Although this current work is a bilingual publication, the Figures have been annotated only in English for the sake of clarity. The graphics, on the other hand, should speak for themselves.

I would like to take this opportunity to thank all our clients who lent us their problems, my colleagues, and last but certainly not least, my wife, all of whom helped bring about this book's success through their understanding and many constructive comments.

Finally, I do sincerely hope that the study of this book will assist the reader in making significantly more efficient use of his HPLC system's capabilities in solving practical problems.

Berlin, February 2003

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