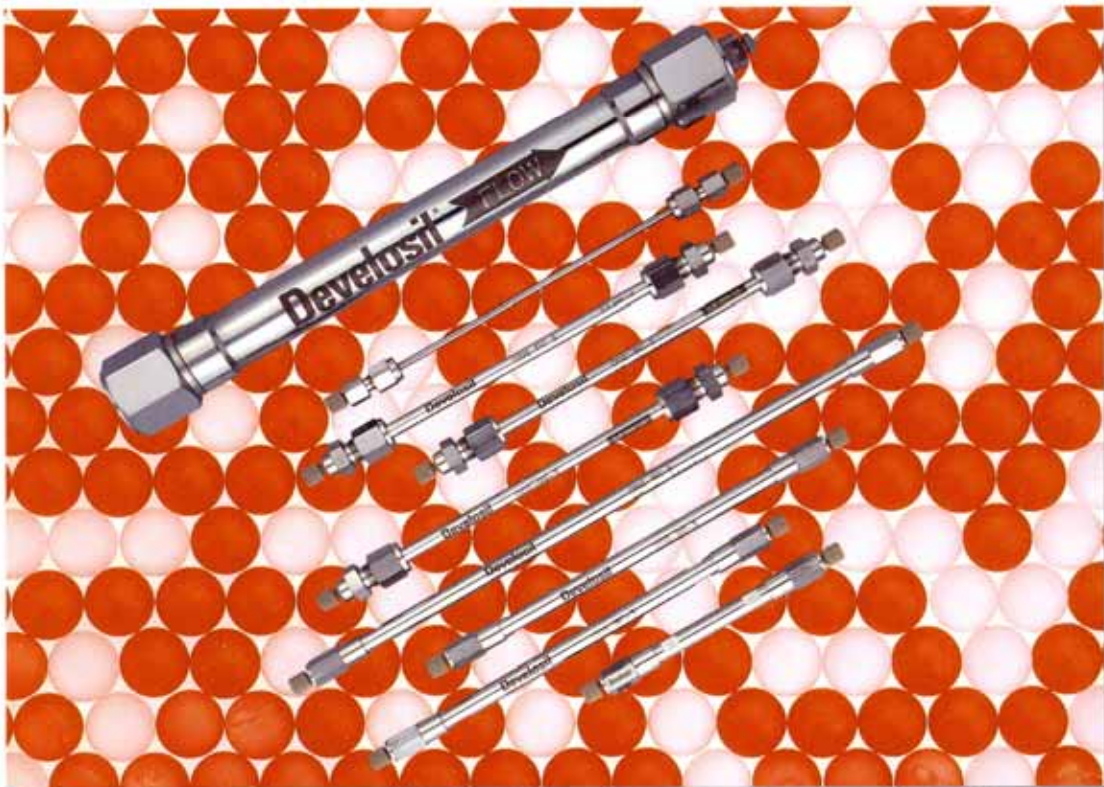


Columns for HPLC

Develosil™



Nomura Chemical Co., Ltd.

Nomura Chemical

Nomura Chemical Co. was founded as a manufacturer of HPLC columns in 1979. We manufacture from silica gel to a final column, and also provide Develosil silica gel or Develosil ODS phases to the other HPLC makers.

We are one of the leading companies of HPLC columns in the world. Especially our patented C30 phase has a quite unique characteristic and has been used in many pharmaceutical corporations, laboratories and universities. Develosil columns are available in the world through our distributors in North America, Europe and Asia.

Develosil HPLC Columns

Columns in 3 kinds of mode such as Reversed, Gel filtration and Normal phase are commercially available.

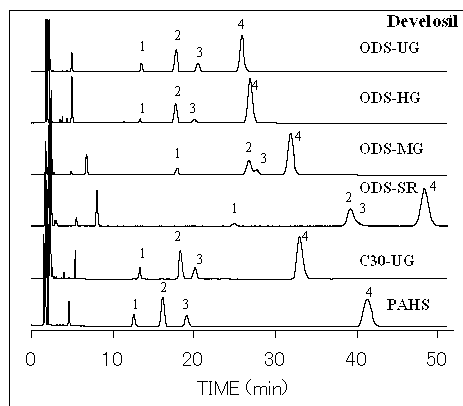
We have 5 kinds of C30 phase, 6 kinds of C18 (ODS) phase and 2 kinds of C8 phase. 300ODS-HG, 300C8-HG and 300C4-HG phases have pores with 25 nm diameter and are for separation of proteins or polypeptides.

And, we have 4 kinds of silica gel. Especially, Develosil 30 (silica gel) has 3 nm pores. Its pore size is the smallest, and it has very large surface area and shows large retention.

MODE	Ligand	PACKING MATERIAL	PARTICLE SIZE
Reversed phase	C30	Develosil C30-UG	3 um, 5 um
		Develosil PRPAQUEOUS	3 um, 5 um
		Develosil Combi-RP	3 um, 5 um
		Develosil RPFULLERENE	3 um, 5 um
		Develosil RPAQUEOUS-AR	3 um, 5 um
	Develosil ERP20	15/30 um	
	C18	Develosil ODS-UG	3 um, 5 um and 15/30 um
		Develosil ODS-HG	3 um, 5 um and 15/30 um
		Develosil ODS-MG	3 um, 5 um and 15/30 um
		Develosil ODS-SR	3 um, 5 um and 15/30 um
		Develosil PAHS	3 um, 5 um
		Develosil 300ODS-HG	5 um
	C8	Develosil C8-UG	3 um, 5 um
Develosil 300C8-HG		5 um	
C4	Develosil 300C4-HG	5 um	
C1	Develosil TMS-UG	3 um, 5 um	
Phenyl	Develosil Ph-UG	3 um, 5 um	
Cyano	Develosil CN-UG	5 um	
Gel filtration phase	Diol	Develosil 300Diol	5 um
		Develosil 100Diol	5 um
Normal phase	Cyano	Develosil CN-UG	5 um
	Amino	Develosil NH2	5 um
	None (Silica)	Develosil 30	3 um, 5 um and 15/30 um
		Develosil 60	3 um, 5 um and 15/30 um
		Develosil 100	3 um, 5 um and 15/30 um
	Develosil SILICA-HILIC(I) and (II)	3 um, 5 um	

Comparison of Develosil ODS (C18) Columns

	ODS-UG	ODS-HG	ODS-MG	ODS-SR	PAHS
Functionarity of C18	Monofunctional	Trifunctional	Difunctional	Difunctional	Trifunctional and polymeric
Ligand density (umol/g)	3.2	3.4	1.6	---	4.5
Carbon content (%)	18	18	15	18	23
Endcapping (TMS)	Yes	Yes	Yes	Yes	No
Pore diameter of silica (nm)	14	14	10	8	12
Surface area of silica (m ² /g)	300	300	450	---	350
Hydrogen bonding capacity k'(caffeine)/k'(phenol)	0.38	0.38	0.48	0.48	0.40
Hydrophobic consistency k'(amylbenzene)/k'(butyl benzene)	1.59	1.58	1.60	1.66	1.58
Steric selectivity k'(triphenylene)/k'(o-terphenyl)	1.50	1.58	1.20	1.21	2.72
Stability	Very good (pH2-10)	Very good (pH1-9)	Good (pH2-7.5)	Good (pH2-7.5)	Good (pH2-7.5)
Retention	Moderate	Moderate	Long (1.3 folds)	Very long (2 folds)	Moderate



Analytical Conditions

Column dimension: 150 x 4.6 mm i.d.
 Mobile phase: Methanol / water = 75 / 25
 Temperature: 30 °C
 Detection: UV at 254 nm
 Sample: 1 = Butylbenzene
 2 = o - Terphenyl
 3 = Amylbenzene
 4 = Triphenylene

The characteristic of each ODS phase is shown in the above table.

Develosil ODS-UG is the most stable under alkaline conditions, and can be used under pH2 - 10.

Develosil ODS-HG is the most stable under acidic conditions (pH1 - 9), and can be used even under 0.5% TFA.

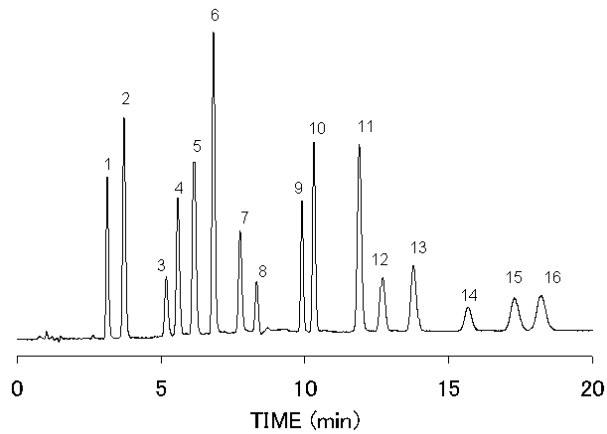
Develosil ODS-MG shows medium performance and suitable for all samples and time to equilibrate is very quick.

Develosil ODS-SR shows the longest retention in our ODS phases, and suitable for LC/MS because much organic solvent obtain high sensitivity.

Develosil PAHS is a real polymeric ODS, and has the highest steric selectivity.

Applications

Separation of Polyaromatichydrocarbons (PAHs)



Column: Develosil **PAHS-5** 150 x 4.6 mm

Mobile phase:

A) Water

B) Methanol

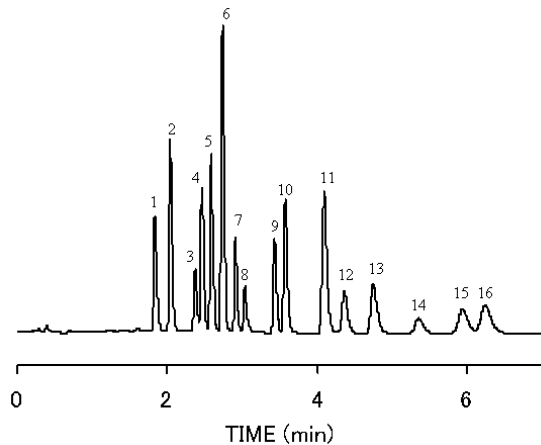
Time	0 min	3.4 min	8.1 min	20min
%B	80%	80%	100%	100%

Flow rate: 1.5 mL/min

Temperature: 30 °C

Detection: UV at 254 nm

Sample: 1 = Naphthalene
 2 = Acenaphthylene
 3 = Acenaphthene
 4 = Fluorene
 5 = Phenanthrene
 6 = Anthracene
 7 = Fluoranthene
 8 = Pyrene
 9 = Benzo [a] anthracene
 10 = Chrysene
 11 = Benzo [b] fluoranthene
 12 = Benzo [k] fluoranthene
 13 = Benzo [a] pyrene
 14 = Dibenzo [a,h] anthracene
 15 = Benzo [g,h,i] perylene
 16 = Indeno [1,2,3-cd] pyrene



Column: Develosil **PAHS-3** 75 x 4.6 mm (3 um particle)

Mobile phase:

A) Water

B) Methanol

Time	0 min	0.5 min	2.5 min	7min
%B	70%	70%	100%	100%

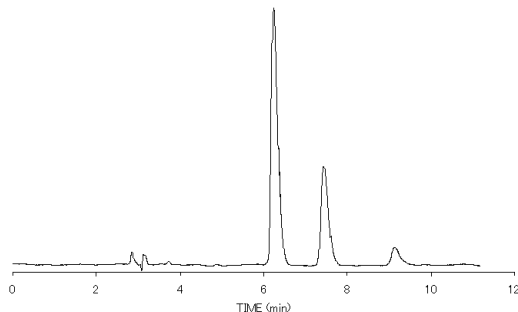
Flow rate: 1.5 mL/min

Temperature: 30 °C

Detection: UV at 254 nm

Sample: 1 = Naphthalene
 2 = Acenaphthylene
 3 = Acenaphthene
 4 = Fluorene
 5 = Phenanthrene
 6 = Anthracene
 7 = Fluoranthene
 8 = Pyrene
 9 = Benzo [a] anthracene
 10 = Chrysene
 11 = Benzo [b] fluoranthene
 12 = Benzo [k] fluoranthene
 13 = Benzo [a] pyrene
 14 = Dibenzo [a,h] anthracene
 15 = Benzo [g,h,i] perylene
 16 = Indeno [1,2,3-cd] pyrene

Separation of Benzalkonium chloride



Column: Develosil **CN-UG-5** 250 x 4.6 mm

Mobile phase: Methanol / 100 mM CH₃COONa, pH5.5 = 70 / 30

Flow rate: 1.0 mL/min

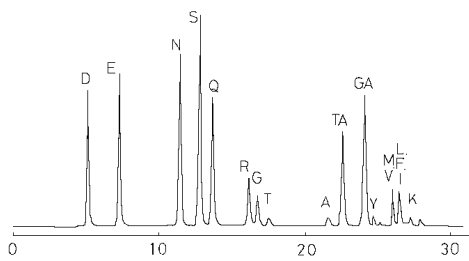
Temperature: 30 °C

Detection: UV at 265 nm

Sample: 1 = benzalkonium chloride

Applications

Separation of Amino acids (OPA)



Data: Tetsuhisa Goto, National Food Research Institute
 Column: Develosil ODS-HG-5 150 x 4.6 mm + 10 x 4.0 mm (guard)
 Mobile phase:

A) 5 mM Citrate buffer (pH6.0) / acetonitrile = 19 / 1
 B) 5 mM Citrate buffer (pH6.0) / acetonitrile = 3 / 7

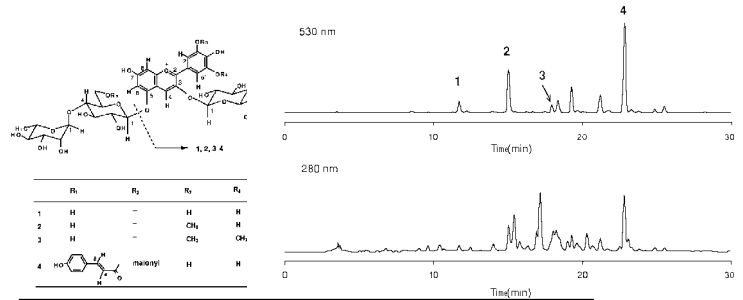
Time	0 min	5 min	20 min	25 min
%B	5%	12%	22%	95%

Flow rate: 1.0 mL/min

Temperature: 40 °C

Detection: Fluorescence Ex. at 340nm, Em. at 450nm

Separation of Anthocyanins



Data: Dr. Kumi Yoshida, Nagoya University
 Column: Develosil ODS-HG-5 250 x 4.6 mm

Mobile phase:

A) 0.5% TFA
 B) TFA / acetonitrile = 0.5 / 99.5

Time	0 min	30 min
%B	10%	30%

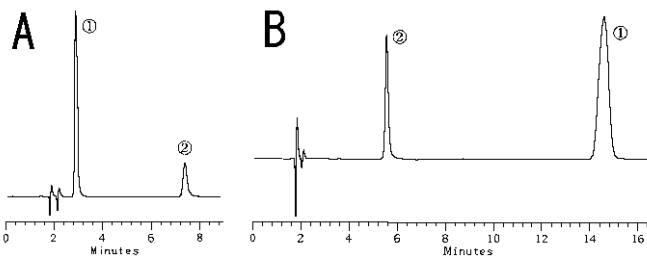
Flow rate: 1.0 mL/min

Temperature: 40 °C

Detection: UV at 530 nm and 280 nm

Sample: Extract of purplish blue spicate flower petal of *Muscari armeniacum*

Separation of Agricultural chemicals 1



Column: Develosil ODS-HG-5 150 x 4.6 mm

Mobile phase:

A) Acetonitrile / 20 mM phosphoric acid = 10 / 90

B) Acetonitrile / 5 mM octansulfonic acid sodium salt + 20 mM phosphoric acid = 10 / 90

Flow rate: 1.0 mL/min

Temperature: 40

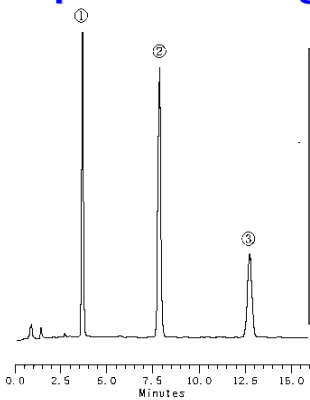
Detection: UV at 250 nm

Sample:

1 = Copper 8-quinolinolate

2 = Methyl sulfanilylcarbamate (Asulam)

Separation of Agricultural chemicals 2



Column: Develosil ODS-HG-5 150 x 4.6 mm

Mobile phase: Acetonitrile / water = 55 / 45

Flow rate: 1.0 mL/min

Temperature: 40 °C

Detection: UV at 230 nm

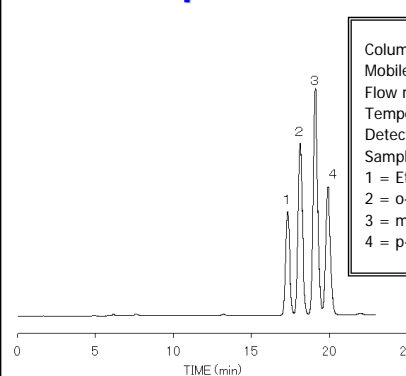
Sample:

1 = Bis(dimethylthiocarbamoyl)disulfide (Thiram)

2 = Iprodione

3 = 0,0-Di-isopropyl S-2-benzensulphonamidoethylphosphorodithioate (Bensulide)

Separation of Xylenes



Column: Develosil PAHS-5 250 x 4.6 mm

Mobile phase: Methanol / water = 75 / 25

Flow rate: 1.0 mL/min

Temperature: 20 °C

Detection: UV at 254 nm

Sample:

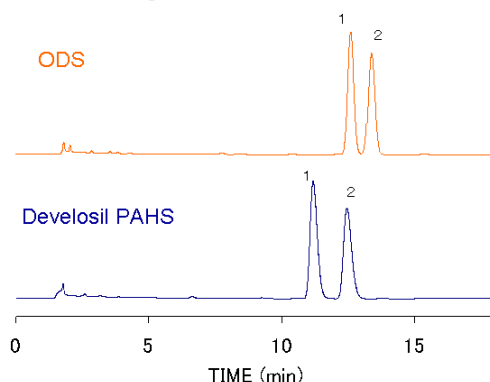
1 = Ethylbenzene

2 = o-Xylene

3 = m-Xylene

4 = p-Xylene

Separation of Vitamin D2 and D3



Analytical Conditions

Column: Develosil PAHS-5 250 x 4.6 mm

ODS 250 x 4.6 mm

Mobile phase: Acetonitrile

Flow rate: 1.0 mL/min

Temperature: 30 °C

Detection: UV at 254 nm

Sample:

1 = Vitamin D2

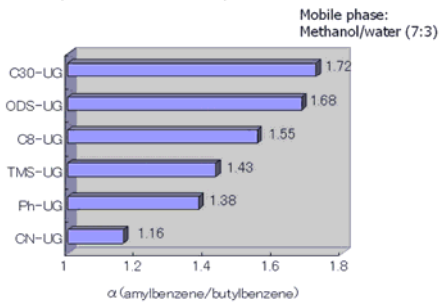
2 = Vitamin D3

UG Series Phases

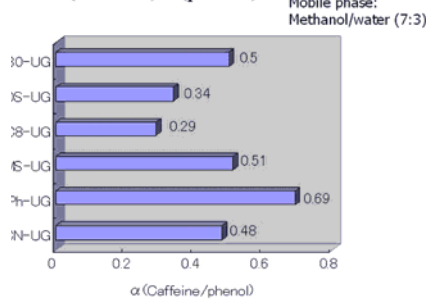
All phase are monomerically bonded and fully endcapped on the same silica base material. Therefore, UG phases can be compared concerning the difference of separation between each ligand.

	Particle size	Ligand	End-capping (TMS)	Carbon content (%)	Silica		
					Surface area (m ² /g)	Pore volume (mL/g)	Pore diameter (nm)
Develosil C30-UG	3 um, 5 um	-Si (CH ₃) ₂ C ₃₀ H ₆₁	Double	18	300	1.15	14
Develosil ODS-UG	3 um, 5 um	-Si (CH ₃) ₂ C ₁₈ H ₃₇	Double	18	300	1.15	14
Develosil C8-UG	3 um, 5 um	-Si (CH ₃) ₂ C ₈ H ₁₇	Double	11	300	1.15	14
Develosil TMS-UG	3 um, 5 um	-Si (CH ₃) ₃	Double	4.5	300	1.15	14
Develosil Ph-UG	3 um, 5 um	-Si (CH ₃) ₂ C ₆ H ₅	Double	8	300	1.15	14
Develosil CN-UG	5 um	-Si (CH ₃) ₂ C ₃ H ₆ CN	Double	7	300	1.15	14

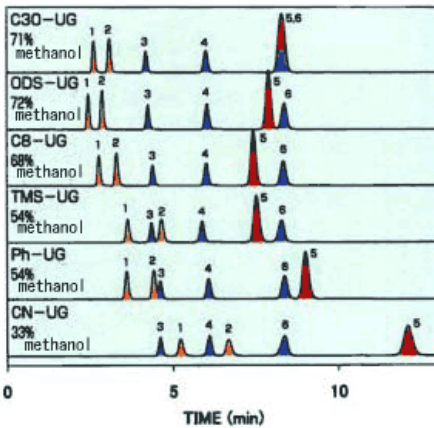
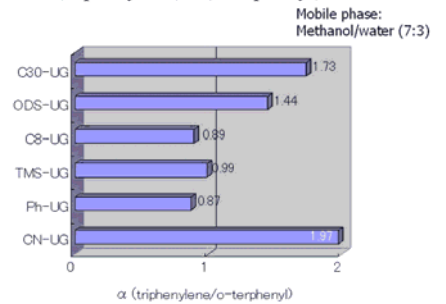
Hydrophobic consistency
k(amylybenzene)/k(butyl benzene)



Hydrogen bonding capacity
k(caffeine)/k(phenol)



Steric selectivity
k(triphenylene)/k(o-terphenyl)



Comparison of chromatograms for standard samples

Conditions
Column size: 150 x 4.6 mm i.d.
Mobile phase: Methanol/ water
(methanol ratio is described in figure) (Retention time of peak No. 6 was adjusted at 8.5 min.)

Flow rate: 1.0 mL/min
Temperature: 30 °C
Detection: UV at 254 nm

Sample
1 = Methyl parabene
2 = Ethyl parabene
3 = Benzene
4 = Toluene
5 = Naphthalene
6 = Ethylbebenze

Develosil Silica Gel

	Particle size	Surface area (m ² /g)	Pore volume (mL/g)	Pore diameter (nm)
Develosil 30	3 um, 5 um, 15/30 um	700	0.5	3
Develosil 60	3 um, 5 um, 15/30 um	500	0.75	6
Develosil 100	3 um, 5 um, 15/30 um	350	1.0	12
Develosil SILICA-HILIC ()	3 um, 5 um	300	1.15	14

Develosil 30, 60 and 100 silica gels are type A silica.

Develosil SILICA-HILIC() is type B and also for HILIC mode.

Expression of Stationary Phases

“Develosil” + stationary phase name (ODS-UG or C8-UG) + particle size (μm)

e.g. Develosil C8-UG-5, Develosil ODS-HG-3, Develosil ODS-UG-5, Develosil C30-UG-5

Dimension of Develosil Columns

Available inner diameters are shown as follows:

Semi-micro column: 1.0 mm i.d., 1.5 mm i.d., 2.0 mm i.d.

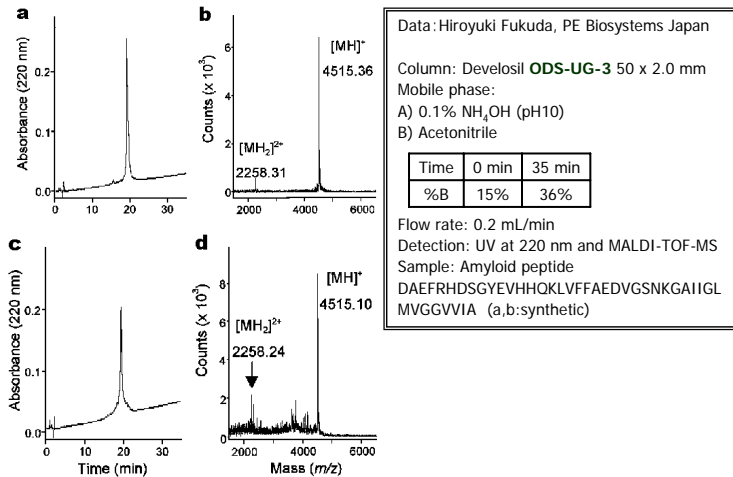
Conventional column: 3.0 mm i.d., 4.0 mm i.d., 4.6 mm i.d., 6.0 mm i.d.

Preparative column: 8.0 mm i.d., 10 mm i.d., 20 mm i.d., 28 mm i.d.

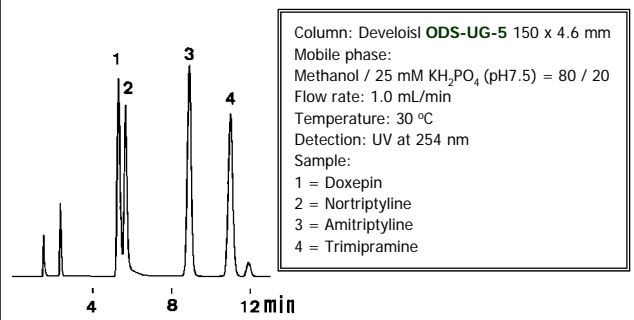
Large scale preparative column: 50 mm i.d. (for >10 or 15/30 um particle)

Applications

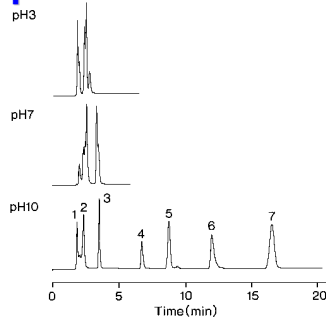
Separation of Amyloid peptide (LC/MS)



Separation of Tricyclic antidepressants



Separation of Basic compounds (effect of pH of a mobile phase)



Column: Develosil ODS-UG-5 150 x 4.6 mm

Mobile phase: Methanol / 20 mM sodium phosphate (pH3.0), (pH7.0), (pH10) = 35 / 65

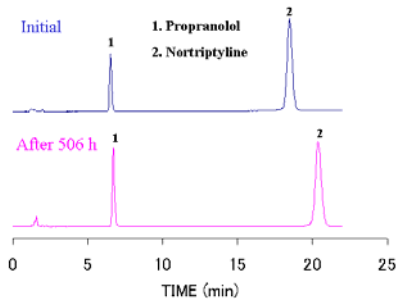
Flow rate: 1.0 mL/min

Temperature: 30 °C

Detection: UV at 254 nm

sample:
1 = Pyridoxine
2 = Amiloride
3 = Pyridine
4 = Benzylamine
5 = Procainamide
6 = N-Methylbenzylamine
7 = N-Acetylprocainamide

Separation of Tricyclic antidepressant (Stability test)



Column: Develosil ODS-UG-5 150 x 4.6 mm + 20 x 4.0 mm guard column

Mobile phase: 0.05M Pyrrolidine-Hydrochloric acid (pH11.5) / acetonitrile = 50 / 50

Flow rate: 1.0 mL/min

Temperature: 30 °C

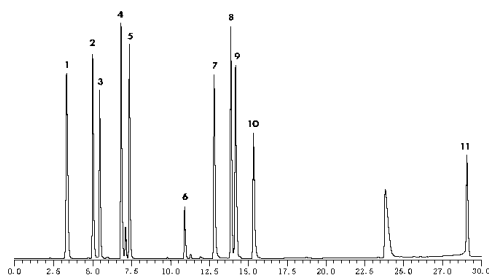
Detection: UV at 215 nm

Sample volume: 2 μ L

	Peak Number	USP Tailing Factor
Initial	1 = Propranolol	1.2
	2 = Nortriptyline	1.1
After 506 hours	1 = Propranolol	1.2
	2 = Nortriptyline	1.2

Peak shape is no change after 506 hours.

Separation of Color additives



Column: Develosil ODS-UG-5 150 x 4.6 mm

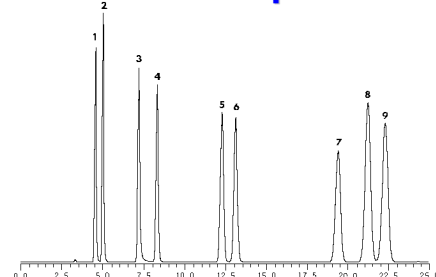
Mobile phase:
A) 10 mM Ammonium acetate (pH6.0)
B) Acetonitrile

Time	0 min	30 min
%B	5%	100%

Flow rate: 1.0 mL/min
Temperature: 40 °C
Detection: UV at 254 nm

sample:
1 = Tartrazine
2 = Amaranth
3 = Indigocarmine
4 = Nwe cocine
5 = Sunset yellow FCF
6 = Fast green
7 = Erythrosine B
8 = Acid red
9 = Phloxine B
10 = Rose bengal
11 = Brilliant green

Separation of Food preservatives



Column: Develosil ODS-UG-5 250 x 4.6 mm

Mobile phase: Acetonitrile / 20 mM sodium acetate (pH4.2) = 40 / 60

Flow rate: 1.0 mL/min

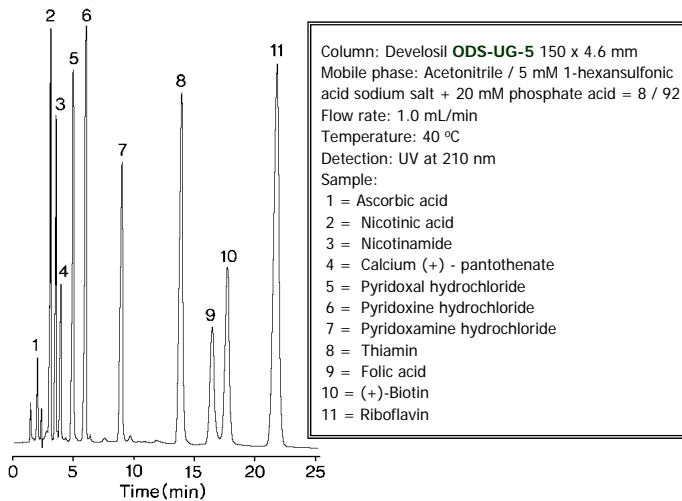
Temperature: 30 °C

Detection: UV at 254 nm

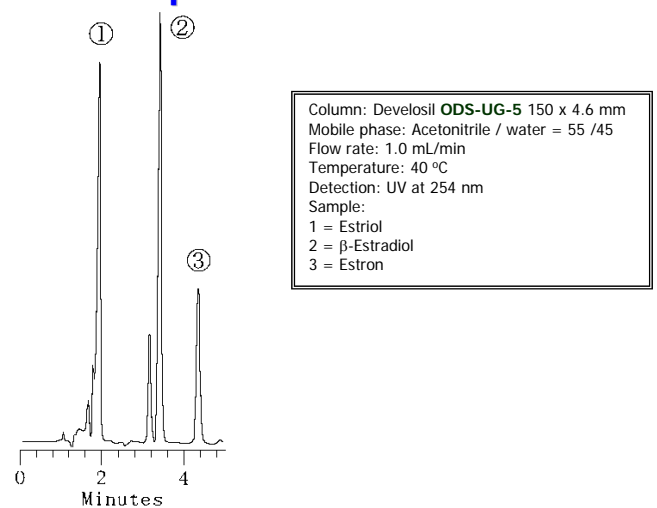
Sample:
1 = Benzoic acid (BA)
2 = Sorbic acid (SOA)
3 = Dehydroacetic acid (DHA)
4 = p-Hydroxybenzoic acid ethyl ester
5 = p-Hydroxybenzoic acid iso-propyl ester
6 = p-Hydroxybenzoic acid n-propyl ester
7 = p-Hydroxybenzoic acid sec-butyl ester
8 = p-Hydroxybenzoic acid iso-butyl ester
9 = p-Hydroxybenzoic acid n-butyl ester

Applications

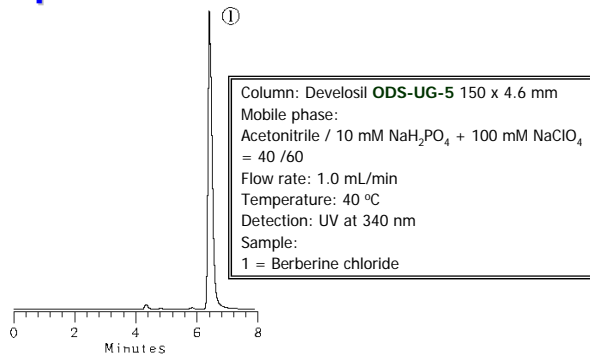
Separation of Water-soluble Vitamins



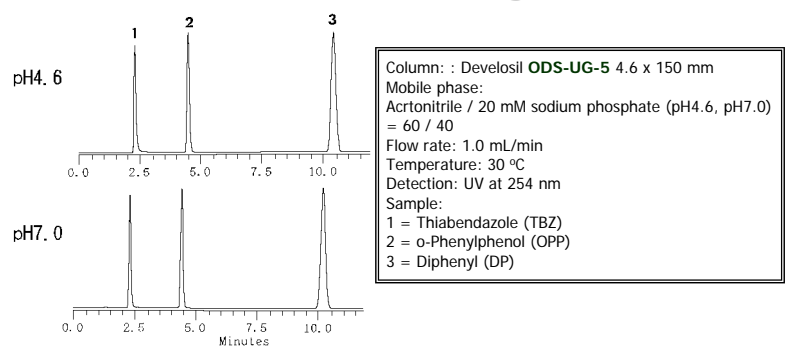
Separation of Steroids



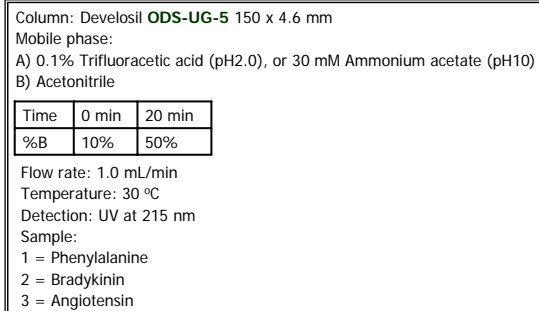
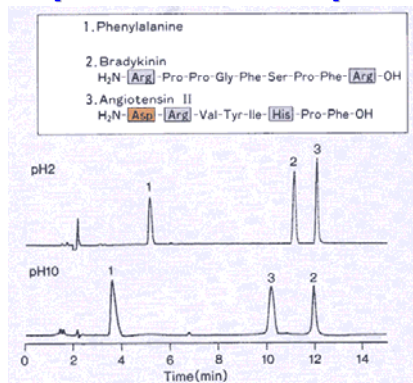
Separation of Berberine chloride



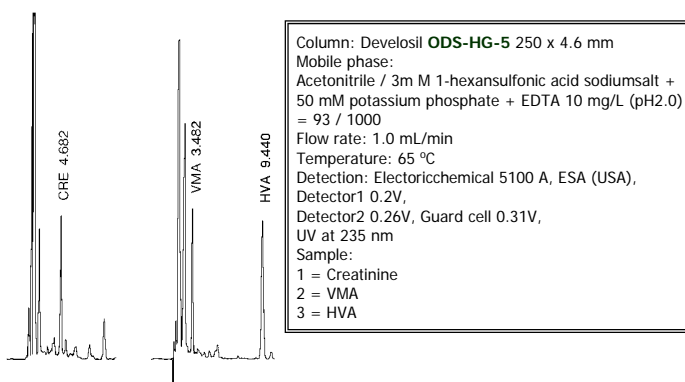
Separation of Fungicides



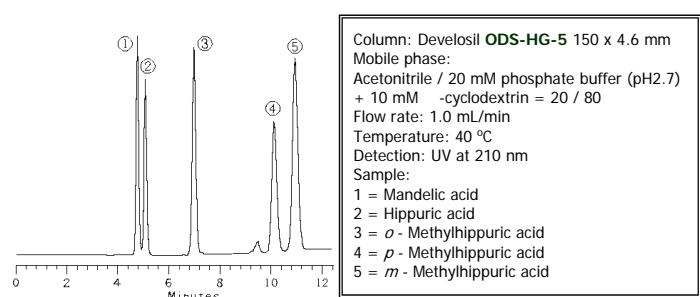
Separation of Peptides (pH effect of mobile phase)



Separation of Creatinine, VMA and HVA



Separation of Hippuric and Methylhippuric acids



Please feel free to contact us "info@develosil.net".

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