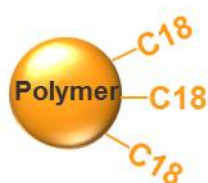


Polymer-based Reversed-Phase C18 HPLC Columns for modern applications: Asahipak ODP

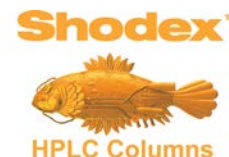
Introduction of polymer RP-C18 columns

Over 60 % of all HPLC applications are Reversed Phase (RP) separations with C18 functional groups. Spherical silica particles proved their use as base material in HPLC columns for many years. To minimize the negative effects of residual silanol groups on the surface, researchers tried end-capping or polymeric coating of the particle and developed hybrid materials. Here we introduce an alternative for these ODS (octadecyl-bonded silica) RP columns:



A polymer particle (for example polyvinyl alcohol) with 4 or 5 μm particle size is modified with C18 alkyl chains as functional groups and used as stationary phase. This HPLC column has greater long term pH stability from 2 to 13. It contains no silanol groups and provides higher resolution for basic substances, for example drugs with tertiary amines (no peak tailing due to unwanted interactions). Also the polarity of the polymer gel is slightly more hydrophilic which gives more retention of polar compounds at the beginning of the chromatogram. The polymer-based RP material can be used in 100 % water or buffer and a variety of organic solvents (isocratic and gradient separation). It has a pore size of 250 Å which makes it suitable for the analysis of proteins and peptides. The very low bleeding is advantageous for the hyphenation to mass spectrometry (MS) or particle-sensitive light scattering detectors. A C18 column with a stationary phase made of polymer offers more than double the life time compared to silica, because it is chemically more resistant.

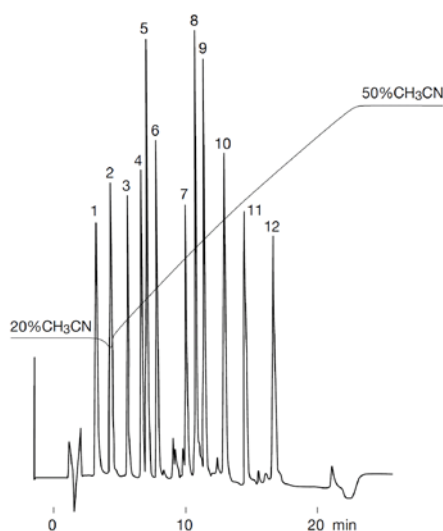
Shodex is specialized in polymer-based packing material for Reversed Phase chromatography. The modern column Asahipak ODP (octadecyl-bonded polymer) combines all the advantages of a polymeric RP-C18 column. It is used for small organic molecules, pharmaceuticals, pesticides, hormones, proteins, vitamins and basic drugs.



Applications on the Asahipak ODP columns

Proteins and peptides

The Asahipak ODP column has a bigger pore size of 250 Å and is ideal for the separation of molecules like proteins and peptides. Aqueous biological buffers can be used with 100 % and organic solvents like acetonitrile or methanol are recommended for flushing. The results show excellent elution volume reproducibility and a high recovery rate.



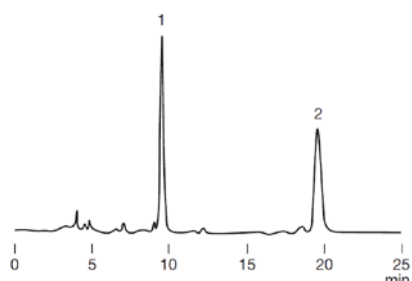
Column: Shodex Asahipak ODP-50 6D
(6.0 x 150 mm, 5 μm)
Eluent: H₂O
Flow rate: 1.0 mL/min
Detection: UV (220 nm)
Temperature: 30° C
Sample: Standard proteins and peptides

Proteins Peptides	Mw	VR(mL) First Analysis	VR(mL) Second Analysis	Recovery (%)
1. Lys-Bradykinin	1188	4.63	4.64	97
2. Bradykinin	1060	5.73	5.70	92
3. Met-Enkephalin	574	7.00	7.00	97
4. Neurotensin	1673	8.04	8.05	99
5. Leu-Enkephalin	556	8.47	8.48	100
6. Substance P	1348	9.24	9.26	93
7. Bacitracin	1450	11.47	11.48	81
8. Insulin	5750	12.24	12.25	95
9. Insulin B chain	3476	12.87	12.89	91
10. Lysozyme	14300	14.43	14.44	96
11. Mastoparan	1479	15.97	15.98	96
12. Myoglobin	17500	18.17	18.18	83

Antibiotics

Due to the increasing use of antibiotics it is important to control its purity during production as well as to monitor residues in clinical or food samples to avoid resistance.

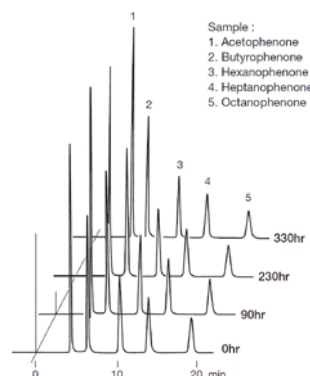
Macrolide antibiotics give the best peak shape under alkaline conditions with pH 11. Erythromycin can be separated from its derivative Azithomycin. Both are on the World Health Organization's list of essential medicines. The polymeric stationary phase of the Asahipak ODP column is long term stable in the range of pH 2 to 13. A smaller particle size of 4 μm was used for better performance.



Column: Shodex Asahipak ODP-40 4E
(4.6 x 250 mm, 4 μm)
Eluent: 40 mM Potassium phosphate
buffer (pH 11.0)/CH₃CN=40/60
Flow rate: 0.5 mL/min
Detection: UV (223 nm)
Temperature: 40° C
Sample: Macrolide antibiotics, 10 μl
1. Erythromycin
2. Azithomycin

Longer life time and stability

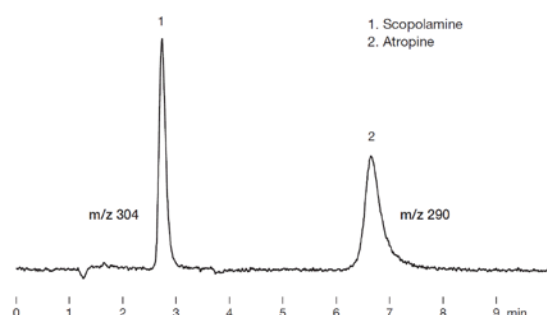
The Asahipak ODP column has a 2.5 times longer life time compared to a silica C18 column. This reduces the down time for replacement and the costs per injection. The durability test with NaOH-solution at pH 12 shows the chemical resistance of the column. The retention time and peak form of the tested phenones are stable and reproducible. 100 % water can be used in gradient or isocratic runs without dewetting effects. This makes the column suitable also for unexperienced users and for method development in R&D for different kind of samples.



Column: Shodex Asahipak ODP-50 4D
(4.6 x 150 mm, 5 μm)
Eluent: 10 mM NaOH aq. (pH 12.0)/CH₃CN=35/65
Flow rate: 0.6 mL/min
Detection: UV (254 nm)
Temperature: 30° C
Sample: Durability test with phenones
1. Acetophenone
2. Butyrophenone
3. Hexanophenone
4. Heptanophenone
5. Octanophenone

Sensitive mass spectrometry detector (MS)

The Asahipak ODP with a smaller inner diameter of 2.0 mm is used for the LC-MS analysis of basic drugs, scopolamine and atropine (both with tertiary amine group). Polymer-based columns exhibit smaller ionic adsorption (no peak tailing) compared to silica-based columns with residual silanol groups. The separation can easily be performed with less or completely without the addition of salt or non-volatile ion pair reagents. Therefore there won't be any unpredictable ion suppression during ionization. The lower bleeding of the Asahipak ODP column results in a better signal-to-noise ratio and makes it also suitable for charged aerosol or particle-sensitive light scattering detectors.



Column: Shodex Asahipak ODP-50 2D
(2.0 x 150 mm, 5 μm)
Eluent: H₂O/CH₃CN=40/60
Flow rate: 0.2 mL/min
Detection: ESI-MS (SIM, positive mode)
Temperature: 30° C
Sample: Basic drugs, 5 μl
1. Scopolamine
2. Atropine

Additional info

Shodex is the brand name of the HPLC columns manufactured by Showa Denko in Japan. They manufacture columns since almost 50 years and produce their own polymer gels and resins for the stationary phases. The product portfolio covers the whole range of HPLC columns, SEC standards and RI detectors. Shodex is a specialist for high-quality polymer-based columns with long lifetime in analytical, micro or preparative dimensions.

Ordering

Product Code	Column Name	Base Material (Polymer)	Functional Group	Particle Size	Pore Size	Column Size (ID x Length)
F7621001	Asahipak ODP-40 4D	Polyvinyl alcohol	C18	4 µm	250 Å	4.6 x 150 mm
F7621002	Asahipak ODP-40 4E	Polyvinyl alcohol	C18	4 µm	250 Å	4.6 x 250 mm
F7620004	Asahipak ODP-50 4D	Polyvinyl alcohol	C18	5 µm	250 Å	4.6 x 150 mm
F7620003	Asahipak ODP-50 4E	Polyvinyl alcohol	C18	5 µm	250 Å	4.6 x 250 mm
F6710022	Asahipak ODP-50G 4A	Polyvinyl alcohol	C18	5 µm	-	4.6 x 10 mm (guard)
F7620002	Asahipak ODP-50 6D	Polyvinyl alcohol	C18	5 µm	250 Å	6.0 x 150 mm
F7620001	Asahipak ODP-50 6E	Polyvinyl alcohol	C18	5 µm	250 Å	6.0 x 250 mm
F6710001	Asahipak ODP-50G 6A	Polyvinyl alcohol	C18	5 µm	-	6.0 x 10 mm (guard)
F7620009	Asahipak ODP-50 2D	Polyvinyl alcohol	C18	5 µm	250 Å	2.0 x 150 mm
F6713001	Asahipak ODP-50G 2A	Polyvinyl alcohol	C18	5 µm	-	2.0 x 10 mm (guard)



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