Vydac BioSelect ® SPE Columns

For Extraction, Concentration and Clean-up of Biological Samples

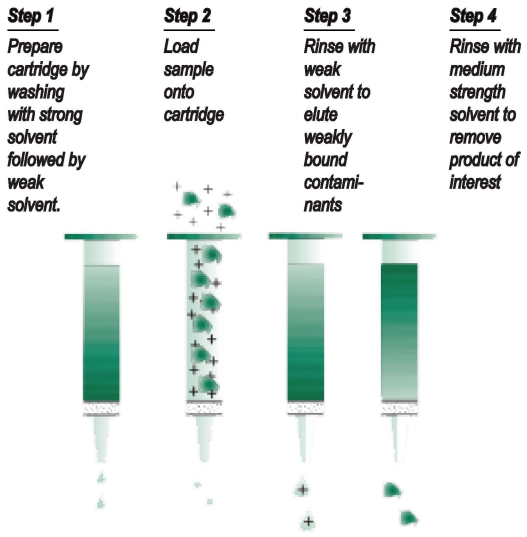
Built using the same high quality media as that of the Vydac HPLC range of columns, the Vydac BioSelect ® SPE columns offer similar selectivity and recovery; rendering it an obvious choice in sample pre-treatment prior to HPLC purification and analysis of biomolecules. Patents referencing the use of Vydac chromatography columns during the biotechnology revolution places the Vydac BioSelect ® chemistries among the most trusted name in biomolecules.

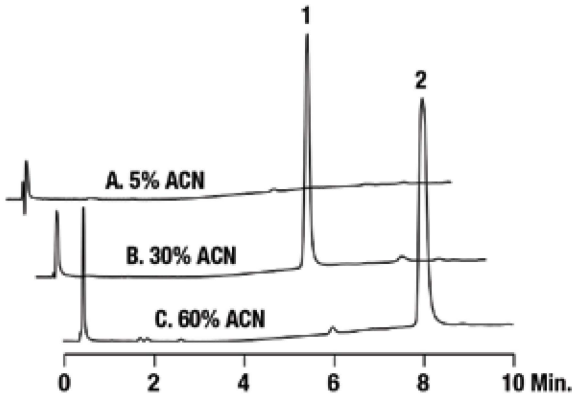
Applications

- Desalting of polypeptide solutions
- Concentration of proteins and peptides
- Removal of HF and cleavage products from cleavage solutions
- Removal of lipids and strongly bound proteins
- Improvement of HPLC resolution by prior removal of early and late eluting by-products or reagents
- Preparation of environment and food samples

Available in C18 and C4 Chemistries

Easy-to-use Vydac Bioselect ™ SPE Columns





Column: Vydac® C4, 5µm, 50 x 4.6mm (Part No. 214TP5405)
Mobile Phase: A: 0.1% Trifluoroacetic Acid B: Acetonitrile
Gradient:

Time:	0	10
%B:	15	70

1. Ribonuclease
2. Myoglobin

Protein Extraction of Ribonuclease and Myoglobin

Procedure using Vydac® SPE:

A 3mL SPE cartridge was conditioned with 1mL of Acetonitrile followed by 1mL of 5% Acetonitrile/ 0.1% Trifluoroacetic Acid. Ribonuclease and myglobin (100mg each) were then loaded in 30% Acetonitrile/ 0.1% Trifluoroacetic Acid . The cartridge was washed with 1mL of 5% Acetonitrile/ 0.1% Trifluoroacetic Acid to remove weakly bound compounds, then 1mL of 30% Acetonitrile/0.1% Trifluoroacetic Acid followed by 1mL of 60% Acetonitrile/0.1% Trifluoroacetic Acid. HPLC analysis of the 5% Acetonitrile wash (A) revealed only a small amount of ribonuclease.Most of the ribonuclease eluted in the 30% Acetonitrile wash (B).The myoglobin eluted almost entirely in the 60% Acetonitrile wash (C).

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Phase	Pore Size (A)	Surface Area (m^2/g)	Carbon Load (%)	End- capped
C18	300	100 m^2/g	8%	Yes
C4	300	100 m^2/g	3%	Yes

Vydac BioSelect® SPE Columns - Ordering Information

Phase	Capacity	Column Size	Pk	Part No.
C4, 13µm	50mg	1ml	50	5103901
	100mg	3ml	50	5103902
C18, 13µm	50mg	1ml	50	5103967
	100mg	3ml	50	5103968
50mg cartridge has 0.5 - 0.75mg polypeptide capacity				
100mg cartridge has 1- 1.5mg polypeptide capacity				

Protocol for Sample Desalting by SPE Prior to Analysis

The SPE step is important for LC-MS analysis. It is not necessary for LC - UV

Reagents and Apparatus All reagents are prepared immediately prior to use. 1% trifluoroacetic acid: Add 100 µL of TFA to 10 mL of water and vortex mix. 0.1 % trifluoroacetic acid: Add 1000 µL of 1% TFA to 10mL of water and vortex mix.

For a 1 mL C18 SPE cartridge (5103967), here is a recommendation for use:

1. Condition cartridge with 1.0 mL of acetonitrile.
2. Rinse with 0.5 mL of water containing 0.1 % TFA. Repeat with another 0.5 mL.
3. Load with 0.2 mL peptide sample containing 0.1 % to 0.2 % TFA for binding.
4. Wash with 0.5 mL of water containing 0.1 % TFA to remove weakly bound components.
5. Elute peptide with 0.2 mL of 75:25 (or up 90:10 acetonitrile:water) containing no TFA.
6. Evaporate off solvent to approximately 10 µL with a stream of nitrogen (or use a vacuum centrifuge with heating no higher than 30 degrees C).
7. Add 190 µL of 5:95 Acetonitrile:Water containing 0.2 % formic acid, 0.01% TFA.
8. Vortex mix and store samples in refrigerator.

Note: To encourage proper fluid flow through the SPE tube, apply positive pressure to the top of tube. This may be accomplished by attaching a 1000 µL pipet tip to a nitrogen gas line; then place the pipet tip over the top opening of the SPE tube.

dr. Wéber Consulting Kft.

www.WEBER.hu

info@weber.hu

Tel: 06 27 330-149