

Product description

for AffiSep® HPLC column kits *

Product description:

AffiSep® HPLC column kits are designed for the analysis, separation and purification of glycoproteins and other glycoconjugates containing lectin specific carbohydrate residues. They combine lectin columns with the corresponding optimised adsorption and elution buffers and column control standards. The kits are for operation with common HPLC or FPLC equipment and facilitate lectin affinity separations of glycoproteins in a simple way for fast and reproducible results.

Kit Contents:

- 1 x AffiSep® HPLC lectin column
- 1 x Adsorption buffer: Dry substance in a plastic bottle
- 1 x Elution buffer: Dry substance in a plastic bottle or ready-to-use
- 1 x Column specificity standard: Lyophilised powder and/ or
- 1 x Column performance standard: Mix of pNP-carbohydrates, 1 mL

The kit composition depends on the choice of lectin.

Column Specifications:

Pore diameter : 1000 Å

Particles: 75 µm mean diameter

Cat.No.	Product description	Lectin	Covalently immobilized lectin [mg/ mL adsorbent]	Amount of bound glycoprotein [µg/ 0.6 mL adsorbent]	Size
031031	AffiSep® WGA kit	<i>Triticum vulgare</i>	~6	≥ 100	0.8 mL
031041	AffiSep® ConA kit	<i>Canavalia ensiformis</i>	~15	≥ 150	0.8 mL
031051	AffiSep® LCH kit	<i>Lens culinaris</i>	~3	≥ 135	0.8 mL
031061	AffiSep® PNA kit	<i>Arachis hypogaea</i>	~3	≥ 125	0.8 mL
031071	AffiSep® AIL kit	<i>Artocarpus integrifolia</i>	~3	≥ 215	0.8 mL
031082	AffiSep® VVL kit	<i>Hairy vetch</i>	~3	-	1.6 mL
031091	AffiSep® AAL kit	<i>Aleuria aurantia</i>	~3	-	0.8 mL
031121	AffiSep® SNA kit	<i>Sambucus nigra</i>	~3	≥ 50	0.8 mL
031131	AffiSep® MAL kit	<i>Maackia amurensis</i>	~3	≥ 125	0.8 mL
031141	AffiSep® UEA kit	<i>Ulex europaeus</i>	~3	-	0.8 mL
031151	AffiSep® GNA kit	<i>Galanthus nivalis</i>	~3	≥ 200	0.8 mL
031161	AffiSep® ECL kit	<i>Erythrina cristagalli</i>	~5	≥ 100	0.8 mL

Please contact for orders and technical support:

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Date: 11.08.2009

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Preparation of buffers and standards:

- Adsorption buffer: The content of the bottle is to be dissolved completely in deionised water like described on the affixed label and will yield buffered saline at 20 °C. Do not adjust pH.
- Elution buffer: The content of the bottle is to be dissolved completely like described on the affixed label and will yield buffered saline containing the required elution sugar at 20 °C. Do not adjust pH.
- For further applications filter the prepared buffer solutions through 0,2 µm.
- Column specificity standard: The lectin-specific glycoprotein is to be dissolved in 1 mL adsorption buffer, inject 100 µL
- Column performance standard: 1 mL carbohydrate standard dissolved in adsorption buffer, inject 100 µL

Storage:

- Add 0,02 % sodium azide to the prepared buffer solutions and store them at 4 °C.
- For long-time storage it is recommended to equilibrate the lectin columns with adsorption buffer containing 0,02 % sodium azide. Columns should be stored at 4 °C.
- Standards should be kept at 4°C.
- Stabilised buffer solutions are stable for at least 6 weeks.

Operational conditions for Lectin columns:

Max. column backpressure: 100 PSI (7 bar)

Recommended flow rate: 0,6 - 0,8 mL/min

Max. flow rate: 1,2 mL/min

Detection wavelength: 280 nm for glycoproteins, 300 nm for p-Nitrophenyl-carbohydrates

Range of pH: AffiSep[®] HPLC lectin columns can be operated between pH 4.0 and pH 8.0



To preserve lectin activity it is recommended to use the supplied buffers containing MnCl₂. Please avoid Mn-precipitation.

Cleaning conditions:

Specifically bound substances can be eluted from the column using the elution buffer. In some cases there might be unwanted unspecific adsorption of contaminants. Then there are the following possible methods for cleaning the lectin columns to remove strongly bound contaminants:

- 2 M NaCl in Adsorption buffer
- 0,5 % Octylglucopyranoside in Adsorption buffer
- 10 % Methanol in Adsorption buffer
- 0,1 % Triton X-100 in Adsorption buffer
- 0,1 M Acetic acid; 0,1 M NaCl; pH 3.0

Apply these cleaning conditions no longer than 20 minutes to the lectin column. After application of one of these steps it is advisable to equilibrate the column with adsorption buffer for at least 1 hour.

* for research, laboratory and in vitro use only

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