# CaptureSelect<sup>™</sup> IgA Affinity Matrix

# INSTRUCTIONS

# Publication Number MAN0009650 Rev. A.0 Introduction

The CaptureSelect<sup>™</sup> IgA Affinity Matrix is designed for the purification of human IgA from recombinant and plasma sources. The affinity matrix contains a 13 kDa single domain [VHH] antibody fragment recognizing all subclasses of human monomeric, dimeric and secretory IgA.

The IgA affinity matrix is suitable for the purification of intact IgA directly from complex source materials in a single step with high purity and yield using neutral pH elution conditions.

The CaptureSelect<sup>™</sup> IgA Affinity Matrix is compatible with FPLC systems.

#### Table 1 Specifications

Ligand	CaptureSelect <sup>™</sup> IgA affinity
Binding specificity	Human IgA from recombinant and plasma sources
Matrix and particle size	Aldehyde-activated agarose, 65 $\mu m$
Dynamic binding capacity	~8 lgA/L of matrix
Shipping solution	20% (v/v) ethanol

#### Table 2 Conditions for use

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Equilibration/wash buffer	20 mM Tris or PBS, pH 7.0 to 7.5	
Elution buffer	0.1 M glycine, pH 3.0	
Flow rate	200 to 400 cm/h	
Pressure limit	≤2 bar	
Cleaning solution	Any of the following:	
	Citric acid or acetic acid	
	<ul> <li>10 to 30 mM NaOH (higher concentrations affect functionality of the affinity ligand on the matrix)</li> <li>PAB (120 mM phosphoric acid, 167 mM acetic acid, and 2.2 %(v/v) benzyl alcohol) prepared freshly every 2 to 3 days and stored protected from light to minimize radicals that affect the functionality of the matrix</li> </ul>	
Storage solution	20% (v/v) ethanol	
	Short term: Room temperature	
	• Long term: 4°C for 2 years	

## Guidelines for use

For optimal matrix performance, optimize the conditions guidelines below for your application.

- Pack the column as described in CaptureSelect<sup>™</sup> Affinity Matrices: Guidelines for Packing (Pub. no. MAN0009645).
- 2. Attach the packed column to the FPLC system.
- 3. Equilibrate with 10 CVs of equilibration/wash buffer.
- **4.** Determine the volume of sample to load based on dynamic binding capacity, concentration of target molecule, and column size. Optimum loading is at physiological pH. Avoid acidic conditions which decrease binding efficiency.
- **5.** Load the sample on the column.
- **6.** Wash with 5 to 10 CVs of equilibration/wash buffer. Washing efficiency can be optimized by increasing NaCl concentration up to 1.0 M.
- 7. Elute with 3 to 5 CVs of elution buffer.
- 8. Re-equilibrate the column in equilibration/wash buffer.
- **9.** Strip the column with 0.1 M Glycine pH 2.0, citric acid, or acetic acid (0.5 to 1.0 M).
- **10.** Re-equilibrate the column in equilibration/wash buffer to prepare the column for another purification run.

## Cleaning

- 1. Pump cleaning solution for 15 minutes.
- **2.** Pump 5 to 10 CVs equilibration/wash buffer to reequilibrate the column.

# **Example application**

Purification of IgA from human serum using CaptureSelect<sup>™</sup> IgA Affinity Matrix

Conditions:

- Column: 0.4 mL CaptureSelect<sup>™</sup> IgA Affinity Matrix, 2 cm bed height
- Equilibration/wash buffer: PBS, pH 7.4
- Load: Human serum, diluted 1:1 in PBS pH 7.4
- Elution buffer: 0.1 M glycine, pH 3.0
- Flow: 200 cm/h

Figure 1 Chromatogram of human IgA purification using CaptureSelect<sup>™</sup> IgA Affinity Matrix – Red line: OD 280 nm signal, blue line: conductivity, gray line: pH



Column: CaptureSelect<sup>™</sup> IgA Affinity Matrix Equilibration/wash buffer: 20 mM Tris, pH 7.4 Load: Human IgA Elution buffer: 20 mM Tris, 2.0 M MgCl2, pH 7.4 Flow: 200 cm/h

**Figure 2** SDS PAGE analysis of the fractions: SYPRO<sup>®</sup> ruby-stained starting material, flow through, and elution fractions of IgA purification using CaptureSelect<sup>M</sup> IgA Affinity Matrix.



- 1: Mol weight marker 2: Starting material
- 3: Flow through
- 4: Elution

# **Ordering information**

CaptureSelect <sup>™</sup> Product	Part Number	
IgA Affinity Matrix	250 mL	1942880250
	500 mL	1942880500
	1 L	19428801L
	5 L	19428805L

# For more information

For more information on CaptureSelect<sup>™</sup> products and ligand leakage ELISA products, go to **www.lifetechnologies.com/ captureselect** 

# Safety information

### **Obtaining SDSs**

Safety Data Sheets (SDSs) are available from **www.lifetechnologies.com/support**.

**Note:** For the SDSs of chemicals not distributed by Life Technologies, contact the chemical manufacturer.

# **Limited Product Warranty**

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