HiTrap IgY Purification HP

Product Information

Cat#No#	Hi-013P

Product Overview

HiTrap IgY Purification HP is a 5 mL column prepacked with Sepharose High Performance for fast and easy purification of IgY from egg yolk.

Description

HiTrap IgY Purification HP is packed with a thiophilic adsorption resin, 2-mercaptopyridine coupled to Sepharose High Performance. Thiophilic adsorption is promoted by water-structuring salts. It has been suggested that the interaction of IgY and ligand results from combined electron donating and accepting action of the ligand or, alternatively, a mixed mode hydrophilic-hydrophobic interaction.

Characteristic

Fast and easy purification of IgY from egg yolk.
Good purity and recovery.
Convenient to use.

Applications

Used for purification of IgY from egg yolk The lipids from an egg yolk were removed by precipitation with water and centrifugation at 4°C.

Maximum operating pressure

5 bar [0.5 MPa] (70 psi)

Sample preparation

As many of the egg yolk lipids as possible must be removed before purification. These can be precipitated using different methods, for example with water or PEG. Precipitation with water is described below.
Separate the egg yolk from the egg white. To one part egg yolk add nine parts of distilled water. Mix and stir slowly for 6 hours at 4°C. Centrifuge at 10 000 × g, at 4°C for 25 minutes to precipitate the lipids. Collect the supernatant containing the IgY.
While stirring slowly, add K2SO4 to the sample to a final concentration of 0.5 M. Adjust the pH to 7.5. Pass

Tel: 1-631-559-9269 1-516-512-3133
Email:info@creative-biomart.com
Fax:1-631-938-8127
45-1 Ramsey Road, Shirley, NY 11967, USA

CREATIVE BIOMART®
Chromatography

HiTrap IgY Purification HP

the sample through a 0.45 µm filter immediately before applying to the HiTrap IgY Purification HP column.

Average particle size
34 µm
Ligand
2-mercaptopyridine
Ligand density
3 mg/ml
Dynamic binding capacity
100 mg pure IgY/column 1/4 egg yolk/column
Recommended flow rate
< 20 ml/min
Recommended column height
25 mm
Temperature stability
4°C to room temperature
Storage
4 to 30°C. 20% Ethanol.
Shipping
20% ethanol
Binding buffer
20 mM sodium phosphate, 0.5 M K2SO4, pH 7.5.
Elution buffer
20 mM sodium phosphate, pH 7.5.
Binding

Tel: 1-631-559-9269 1-516-512-3133

Email:info@creative-biomart.com

Fax:1-631-938-8127

45-1 Ramsey Road, Shirley, NY 11967, USA

HiTrap IgY Purification HP

To improve recovery of total IgY or a specific IgY antibody the 0.5 M K_2SO_4 in the binding buffer can be replaced with 0.6 to 0.8 M Na_2SO_4. The sample should have the same concentration of Na_2SO_4 as the binding buffer. An increase in salt concentration will, however, adversely affect the purity of the eluted IgY.

Elution

The purity of the eluted IgY may be improved by using gradient elution with, for example, a linear gradient 0–100% elution buffer over 10 column volumes, followed by elution at 100% elution buffer for a few column volumes.

Cleaning-in-place

20 mM sodium phosphate, pH 7.5 with 30% isopropanol.

Purification procedures

1. Remove the stopper.
2. Fill the syringe or pump tubing with binding buffer. Using the connector provided, connect the column “drop to drop” to the syringe, or pump tubing, to avoid introducing air into the column.
3. Remove the snap-off end at the column outlet.
4. Wash the column with at least 5 column volumes (CV) of each buffer: Binding, elution and cleaning buffers.
5. Equilibrate the column with 5 CV of binding buffer.
6. Apply the sample, using a syringe fitted to the luer adapter or by pumping it onto the column.
7. Wash with at least 10 CV of binding buffer or until no material appears in the effluent.
8. Elute the IgY with 10 CV of elution buffer.
9. Regenerate the column with 8 CV of cleaning buffer.
10. Re-equilibrate the column with 5 CV of binding buffer.

Pack size

5 mL

Maximum flow velocity

20 ml/min

Dimensions

16 × 25 mm

Tel: 1-631-559-9269 1-516-512-3133

Email:info@creative-biomart.com

Fax:1-631-938-8127

45-1 Ramsey Road, Shirley, NY 11967, USA

CREATIVE BIOMART®
Chromatography

HiTrap IgY Purification HP

Column volume
5 ml
Column i.d.
16 mm
Column hardware pressure limit
5 bar (0.5 MPa, 70 psi)

Tel: 1-631-559-9269 1-516-512-3133

Email:info@creative-biomart.com

Fax:1-631-938-8127

45-1 Ramsey Road, Shirley, NY 11967, USA