

GammaBind Plus Sepharose antibody purification resin

Product Information

Cat#No#	Ga-326C
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Product Overview

GammaBind Plus Sepharose is a protein G affinity chromatography resin for purifying immunoglobulins (IgGs) from a variety of species: Enhanced binding to rat and mouse IgG subclasses compared with GammaBind G Sepharose.

Binds to all mouse, rat, and human IgG subclasses; binds total IgG from guinea pig, rabbit, goat, cow, sheep, and horse.

Lab-scale IgG purification yields up to 35 mg human IgG/mL.

Well suited for immunoprecipitation procedures.

Description

GammaBind Plus Sepharose is GammaBind G, Type 3, covalently immobilized to Sepharose CL-6B by malimide linkage. This rigid matrix results in easy handling and fast separations.

GammaBind G, Type 3, a recombinant form of streptococcal protein G, binds to the Fc region of IgG from a variety of mammalian species. GammaBind Plus Sepharose may be used to analyze and purify classes, subclasses and fragments of immunoglobulins from any biological fluid or cell culture medium. Since only the Fc region is involved in binding, the Fab region is still available for binding antigen. Hence, GammaBind Plus Sepharose is very useful for isolation of immune complexes

Applications

GammaBind Plus Sepharose is used to analyze and purify classes, subclasses, and fragments of immunoglobulins from any biological fluid or cell culture medium. This resin is also suitable for immunoprecipitation.

Medium Preparation

GammaBind Plus Sepharose is supplied preswollen in phosphate buffered saline (PBS), pH 7.0 containing 20% ethanol as preservative. Prepare a slurry by decanting the phosphate buffered saline solution and replace it with binding buffer, in a ratio of 75% settled medium to 25% buffer. The binding buffer should not contain agents which significantly increase the viscosity. The column may be equilibrated with viscous buffers at reduced flow rate after packing is completed.

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For batch procedures remove the phosphate buffered saline solution by washing the medium on a medium porosity sintered glass funnel.

Ligand Coupling Method

Maleimide activation

Matrix

Cross-linked agarose, 6%

Average particle size

~90 µm

Ligand

Recombinant GammaBind G type 3 lacking albumin-binding region

Ligand density

3 mg GammaBind G, type 3/ml medium

Coupling chemistry

maleimide linkage

Dynamic binding capacity

35 mg human IgG/ml drained medium

7 mg mouse IgG/ml drained medium

Recommended flow rate

< 130 cm/h

Chemical stability

Stable to all commonly used aqueous buffers and additives such as 1 M acetic acid, 1% SDS and 6 M guanidine hydrochloride.

Physical stability

Negligible volume variation due to changes in pH or ionic strength.

pH working range

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3–9

pH CIP range

2–9

Storage

2 to 8°C, 20% Ethanol

Notes

If you have packed at the maximum flow velocity, do not exceed 75% of this in subsequent chromatographic procedures.

Binding buffer

0.01 M sodium phosphate buffer, 0.15 M NaCl, 0.01 M EDTA, pH 7.0.

Elution buffer

0.5 M acetic acid, pH 3.0.

Binding

IgG from most species binds to GammaBind Plus Sepharose at neutral pH and physiological ionic strength. As a general method we recommend 0.01 M sodium phosphate, 0.15 M NaCl, 0.01 M EDTA, pH 7.0 as binding buffer.

Adjust the pH of the sample before it is applied to the column, either by buffer exchange on a HiTrap Desalting column, PD-10 column or HiPrep 26/10 Desalting column depending on the sample volume. The binding capacity of GammaBind Plus Sepharose depends on the source of the particular immunoglobulin. However, the total capacity depends upon several factors, such as the flow rate during sample application, the sample concentration and binding buffer.

Equilibration

Equilibrate all material to the temperature at which the chromatography will be performed.

Elution

Bound antibodies can be eluted with high yields over a pH range from 2.5 to 3.0.

As a general method, we recommend 0.5 M acetic acid adjusted to pH 3.0 with ammonium hydroxide.

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As a safety measure to preserve the activity of acid labile IgG's, we recommend the addition of 60 to 200 µl 1 M Tris-HCl, pH 9.0 per ml eluted fraction, for neutralization of the eluted fractions.

Regeneration

After elution, the medium should be washed with 2 to 3 column volumes of cleaning buffer, 1 M acetic acid, pH 2.5, followed by reequilibration with 2 to 3 column volumes of binding buffer.

Cleaning-in-place

pH stability where the medium can be subjected to cleaning- or sanitization-in-place without significant change in function.

Pack size

5 mL

Maximum flow velocity

130 cm/h

Maximum operating backpressure

0.015 MPa (0.15 bar, 2 psi)
