

PRODUCT DATA SHEET



Bioepitope[®] Protein A Agarose

Catalog Number: BD0043

Quantity: 8ml

Description: Protein A is provided as agarose conjugate for use in antibody affinity purification. It also can be used for immunoprecipitation, just dilute the agarose 2 times with PBS. The product is provided as 4 ml agarose in 4 ml PBS. Protein A-agarose is pre-blocked with BSA to reduce non-specific immunoglobulin binding.

Specificity: Protein A-agarose is suitable for antibody affinity purification of mouse IgG2a, IgG2b, and IgA, rabbit IgG, and human IgG1, IgG2 and IgG4.

Format: PBS, 4ml agarose

Storage: Store at 4° C, do not freeze; stable for one year from the date of shipment.

Procedure:

Sample pre-treatment: Check the pH of the sample, and adjust, if necessary, before applying the sample to the column. The pH of the sample should be equal to the pH of the binding buffer. pH can be adjusted by either diluting the sample with binding buffer or by buffer exchange using desalting columns or dialysis. Clarify the sample before applying it to the medium.

Binding buffer: 20 mM sodium phosphate, pH 7.4;

Elution buffer: 0.1 M glycine-HCl, pH 2.8;

Neutralizing buffer: 1 M Tris-HCl, pH 8.5;

This protocol is suitable for gravity-flow chromatograph.

1. Equilibrate the resin and all other buffers to room temperature. Cut off the bottom tip and remove the top cap. Stir or swirl bottle to evenly suspend the resin, and then transfer the resin to the column. Equilibrate the column with 10 ml of binding buffer.
2. After equilibration, add the sample. A volume of 1 to 10 ml is recommended. If the sample volume is less than 1 ml, dilute to 1 ml with binding buffer.
3. Apply the diluted sample to the column and allow it to flow completely into the resin. Do not allow the resin bed to run dry. Any volume may be applied provided the total amount of antibody is less than 80% of column capacity.
4. Wash the column with 15mL of the Binding Buffer.
5. Elute antibodies with 5mL of Elution Buffer and collect 0.5-1mL fractions. Immediately adjust eluted fractions to physiologic pH by adding 100µL of the Neutralization Buffer per 1mL of eluate. Monitor the elution by measuring the absorbance at 280nm or by protein assay such as Bioworld BCA

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Protein Assay Kit (BD0028).

6. Pool the eluted IgG fractions that contain the high absorbance. The purified antibodies may be used directly for SDS-PAGE, or the buffer may be exchanged by dialysis or desalting column to one that is compatible with the specific downstream application.
7. Regenerate column by washing with 10mL of Elution Buffer. Columns may be regenerated at least 10 times without loss of binding capacity. And wash it with 5 to 10ml of binding buffer. For storage, wash column with 5-10mL of water containing 0.02% sodium azide. When approximately 3mL of solution remains, replace the bottom cap followed by the top cap on the column. Store columns upright at 4°C.

Research Use: For research use only, do not for use in diagnostic procedures.

Ordering information:

Product	Specificity	Catalog Number	Amount
Protein A Agarose	mouse IgG2a, IgG2b and IgA rabbit polyclonal Abs human IgG1, IgG2 and IgG4	BD0043	8ml
Protein G Agarose	mouse IgG1, IgG2a, IgG2b and IgG3 rat IgG1, IgG2a, IgG2b and IgG2c rabbit and goat polyclonal Abs human IgG1, IgG2, IgG3 and IgG4	BD0044	8ml
Protein A+G Agarose	all of the above Abs	BD0045	8ml

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