Antibody Purification Kit (W/O Column)

P1456508

Storage 2-8℃.

Shipping Shipped with ice packs. Please store under the specified storage conditions immediately upon receipt.

Introduction

The purification kit is a highly efficient and flexible tool specifically designed for high-purity antibody purification from a variety of sample sources (such as serum, ascites, cell culture supernatant, or hybridoma supernatant). Unlike traditional pre-packed column kits, this kit does not include pre-packed columns or chromatography media. Instead, you can select and pack suitable media into empty columns of any scale according to your experimental needs, making it perfectly adaptable for both manual operation and automated purification systems. This product provides a high-concentration Protein A/G purification buffer, offering researchers maximum experimental flexibility to accommodate various empty chromatography columns, gravity-flow columns, or batch purification processes.

Product Components

P1456508	Component	20 T	50 T	Storage	Quantity Per Test
P1456508A	Binding buffer (10×)	50 mL	250 mL	2-8℃	5 mL
P1456508B	Elution buffer (5×)	10 mL	50 mL	2-8℃	1 mL
P1456508C	Neutralizer Buffer	10 mL	50 mL	2-8℃	1 mL
P1456508D	Coomassie Brilliant Blue Staining Solution	10 mL	50 mL	2-8℃	1 mL
P1456508E	Regeneration Buffer	10 mL	50 mL	2-8℃	10mL (Optional)

Note: The recommended amount of Purification resin for each test is 2 mL.

Instruction for use

Prior to use, dilute all component buffers to 1× concentration with deionized water. Specific operations can be adjusted according to the requirements of different purification tools.

I. Sample Preparation

Before purification, clarify hybridoma supernatants, serum, or ascites samples by centrifugation or filtration through a 0.45 μm filter to avoid clogging the purification column and reducing its lifespan.

II. Sample Purification

1. Equilibration

Fill the purification column with 1× Binding Buffer (approximately 15 column volumes) to equilibrate the column, allowing the liquid to drain naturally.

2. Sample Loading

Remove the top cap of the purification column and add the clarified sample to the purification column, allowing the liquid to drain naturally.

Note: If the antibody concentration in the sample is low or the sample volume is large, increase the number of loading cycles or extend the loading time to improve the binding efficiency between the antibody and the resin.

3. Washing

Fill the purification column with 1× Binding Buffer (approximately 15 column volumes) to wash away unbound impurities, allowing the liquid to drain naturally.

4. Elution

4.1 Prepare several 1.5 mL centrifuge tubes and pre-add 10 μL of Neutralizer Buffer to each tube.

4.2 Add 3 mL of 1× Elution Buffer (approximately 3 column volumes) to each purification column and collect the eluate by gravity. Collect 8 drops (approximately 0.5 mL) per tube and repeat until elution is complete.

5. Dialysis

5.1 Take 2.5 μL of eluate from each tube, mix with 10 μL of Coomassie Brilliant Blue Staining Solution, and observe the gradient change of antibody elution.

5.2 Collect the eluate tubes with high antibody concentration and dialyze against an appropriate buffer to obtain highly purified IgG.

III. Storage of Purification Column

1. After antibody elution, add an additional 2 mL of 1× Elution Buffer to the column to wash away any residual antibody.
2. Fill the purification column with 1× Binding Buffer (approximately 15 column volumes) to equilibrate the column, allowing the liquid to drain naturally.
3. Fill the purification column with 20% ethanol (approximately 15 column volumes) for preservation, allowing the liquid to drain naturally.
4. Add 5 mL of 20% ethanol to the column, cap the top, and seal the bottom cap. Store at 2-8℃ for direct use in subsequent purifications.

IV. Column Regeneration (Optional)

Perform regeneration when a measurable decrease in column performance is observed, following these steps:

1. Add 10 mL of Regeneration Buffer to the column for washing.
2. Add 30 mL of double-distilled water (ddH_2O) to rinse the column thoroughly and remove

residual Regeneration Buffer.

3. Add 20 mL of 1× Binding Buffer to wash and equilibrate the resin.
4. Add 15 mL of 20% ethanol for washing, then add 5 mL of 20% ethanol to the column. Cap the top and seal the bottom cap, and store at 2-8℃.