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Anti-Rabbit Beads

Catalog # B0003-5
Size 5ml

Introduction

Anti-Rabbit Beads contains sepharose beads coated with goat anti-rabbit IgG, for purification of rabbit IgG class of antibodies or proteins with affinity to goat anti-rabbit IgG. The goat anti-rabbit IgG is affinity purified and conjugated to the beads at 1-1.5 mg/ml ratio. This product can be used for 100-200 times. For frequent use, an aliquot can be stored at 4ºC for 1 month with addition of 0.02% sodium azide (NaN3) to the storage buffer. It may also be used for immunoprecipitation (IP).

Note: 20% ethanol was contained as protection solution in this product, please wipe off the ethanol before use.

Anti-Rabbit Beads Specifications

Matrix: CNBr-activated Sepharose^{TM} 4FF
Beads concentration: 1-1.5 mg/ml
Coupling conditions of matrix: pH 7-9, 4°C to 25°C, 2-16 h
Binding capacity: 4-7 mg IgG per ml
Bead size range: 45–165 μm
Mean bead size: 90 μm
Bead structure: Highly cross-linked agarose, 4%
Max. flow rate: 4 ml/min/cm^2
Recommended flow rate: 1-3 ml/min/cm^2
Stability of the matrix: pH 3-11 (ligand dependent)
Storage: Store at 4℃ for frequent use, at -20℃ for at least one year.

Protocol

A: Buffers preparation

- Equilibration buffer A: 1% Nacl+0.1% Na_2HPO_4, pH≈7.5
- Equilibration buffer B: 1% CH3COONa adjusted pH to 5 by CH_3COOH.
- Elution buffer: CH_3COOH(pH =2~3) or 0.1mol Glycine Hydrochloride..
- Wash buffer: 1% Nacl+0.1% Na_2HPO_4, pH≈7.5
- Storage buffer: 30% glycerol

B. Sample preparation

1. Dilute the serum with equilibration buffer A to ensure its content and pH closed to equilibration buffer A.
2. Centrifuge diluted serum supernatants to sediment debris.
3. Filter supernatants through 0.45μm filter.

C. Affinity-purification

1. Load the Anti-rabbit beads into the empty column.
2. Wash column with Wash buffer in 3-5 column volumes to remove the glycerol, and then, equilibrate column by washing with Equilibration buffer A in 5-10 column volumes.
3. Bring the sample to room temperature, and load it into the column by a syringe or a pump. The total volume of the sample applied is not critical in most cases.
4. Load the sample into the column and collect the flow liquid, repeat this action for 3-5 times. If necessary, repeat for more times, then deal with the collected liquid reasonably.
5. Wash the column with Equilibration buffer B to remove other proteins.
6. Elute with Elution buffer, collect the flow liquid (antibody), adjust its pH by saturated Na_2CO_3 during collection. Then, customers can test the related data of the antibody as their own requirements.

D. Re-equilibration and Storage

1. Add 5-10ml Elution buffer to column to elute thoroughly, then neutralizate the column with Equilibration buffer A.
2. Wash the column bed with Storage buffer in 3-5 column volumes, seal the bottom of the column and store at -20℃ for at least one year. For frequent use, an aliquot can be stored at 4ºC for 1 month with addition of 0.02% sodium azide (NaN_3) to the storage buffer.