

Protein A/G-Sepharose Column

CATALOG #: 6528-1 1 ml
6528-5 5 ml

LOT #: _____

PREPARATION: Protein A/G Sepharose is prepared by covalently coupling recombinant Protein A/G (contains five Ig-binding regions of protein A and three Ig-binding regions of protein G, Cat. 6502) to 6% cross-linked sepharose beads. The coupling was optimized to give a high binding capacity for IgG. The capacity of IgG binding could be greater than 10 mg of human IgG per ml of wet bead.

CONTENTS: Ready-to-use pre-packed columns of 1 ml or 5 ml bead volume in PBS with 0.02% sodium azide.

FEATURES: Binding capacity of human IgG greater than 10 mg/ml of bead; High flow rate; Low falling off of rProtein A/G; pH stability 2-10.

APPLICATIONS: Purification of monoclonal and polyclonal antibodies.

STORAGE: Store at 4°C. Do not freeze. Stable, as supplied, for at least 1 year.

PROCEDURE EXAMPLE:

1. Equilibrate the column to room temperature.
2. Remove the upper (first) then lower cap and allow the preservative to drain by gravity flow.
3. Equilibrate the column with 5 – 10 bed volumes of degassed Binding Buffer.
4. Add sample in Binding Buffer and recycle through column 15-20 times.
5. Wash with 4 – 5 column volumes of Binding Buffer containing 0.5 M NaCl
6. Wash with at least 2 column volumes of Binding Buffer and ensure the effluent reaches the same Absorbance (280 nm) as the Binding Buffer.
7. Drain the column to the top frit. Elute with one bed volume of Elution Buffer buffer pH 3.0. Neutralize with 100 µl of 1M Tris, pH 9.0 per ml of eluate. It is recommended to elute with another 1-2 ml of elution buffer and collect 100 µl fractions (test for absorbance at 280 nm).
8. Alternatively, one could add 6 ml Elution Buffer and collect 1 ml fractions into 1.5 ml tubes containing 100 µl of 1M Tris, pH 9.0.
9. Combine fractions with highest absorbance (Remember to blank the spectrophotometer with a solution containing 100 µl 1M Tris, pH 9.0 per ml of Elution Buffer.
Concentration of IgG (mg/ml) = (A280/1.38)
10. Regenerate the column by:
 - a. Washing with ~ 5 volumes of Elution Buffer.
 - b. Equilibrate with 5 volumes of Binding Buffer containing 0.2% sodium azide.
 - c. Store upright at 4 °C.

BUFFER EXAMPLES:

- (1) **Binding Buffers:** 50 mM sodium borate, 0.15M sodium chloride pH 8.0.
- (2) **Elution Buffers:** 0.1 M citric acid, pH 2.75

RELATED PRODUCTS:

Recombinant Protein G- Sepharose Column	Protein G Polyclonal Antibody
Recombinant Protein L- Sepharose Column	Protein A Polyclonal Antibody
Recombinant Protein A- Sepharose Column	Protein L Polyclonal Antibody
Recombinant Protein A/G/L- Sepharose Column	Protein G Magentic Beads
Recombinant Protein A Sepharose Beads	Protein A Magnetic Beads
Recombinant Protein G Sepharose Beads	Protein L Magnetic Beads
Recombinant Protein L Sepharose Beads	Protein A/G Magentic Beads
Recombinant Protein A/G Sepharose Beads	Protein A/G/L Magnetic Beads
Recombinant Protein A/G/L Sepharose Beads	

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