rev 07/11

For research use only

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.

USAGE: For Research Purpose Only! Not to be used in humans!

Procedure Example:

- 1. Equilibrate the column to room temperature.
- 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 μ l 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 μ l 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 μ l of 1M Tris, pH 9.0.
- 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/mI) = (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