rev. 03/12

Protein G-Sepharose Column

CATALOG #: 6518-1 1 ml

6518-5 5 ml

LOT #: _____

PREPARATION: Protein G Sepharose is prepared by covalently coupling

Recombinant Protein G (contains three IgG binding domain, BV catalog # 6510-10) to 6% cross-linked sepharose beads. The coupling technique is 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 20 % Ethanol/dH2O.

FEATURES: Binding capacity of human IgG greater than 10 mg/ml of

bead; High flow rate; Low falling off of rProtein G; pH stability

2-10.

 $\textbf{Note:} \ \, \text{Protein} \ \, \text{G} \ \, \text{binds to all IgG subclasses from human},$

mouse and rat species. It also binds to total IgG from guinea

pig, rabbit, goat, cow, sheep, and horse.

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 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).
- 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.
- 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 20% ethanol in water.
 - Store upright at 4 °C.

BUFFER EXAMPLES:

(1) Binding Buffers: 25 mM sodium phosphate pH 7.2, 50 mM Tris, pH 7.2 or PBS, pH 7.2

(2) Elution Buffers: 0.1 M glycine, pH 3.0 or 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/G- Sepharose Column Protein L Polyclonal Antibody Recombinant Protein A/G/L- Sepharose Column Protein G Magnetic 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 Magnetic Beads Recombinant Protein A/G Sepharose Beads Protein A/G/L Magnetic Beads

Recombinant Protein A/G/L Sepharose Beads

FOR RESEARCH USE ONLY! Not to be used on humans.

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