rev. 08/14 For research use only

## **Protein L-Sepharose Column**

**CATALOG #**: 6538-1 1 ml

6538-5 5 ml

LOT #: \_\_\_\_\_

PREPARATION: Protein L-Sepharose is prepared by covalently coupling

recombinant Protein L (contains five Ig light chain binding domains, BV catalog # 6530-10) to 6% cross-linked sepharose beads. The coupling technique is optimized to give a high binding capacity. The capacity of IgG binding is generally 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/H<sub>2</sub>O.

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

bead; High flow rate; Low falling off of rProtein L; 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:

- 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  $\mu$ 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  $\mu$ 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  $\mu$ l 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/mI) = (A280/1.38)

- 10. Regenerate the column by:
  - 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:**

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 A/G- 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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