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# TECHNICAL INFORMATION

## ImmunO™

Catalog Number: 987051
**Recombinant Protein A**

**Description:** Recombinant protein A binds immunoglobulins (primarily IgG) at the Fc region with high affinity. In some cases, IgM and IgA will also bind. This property provides a rapid, simple and economical method for purification and analysis of a broad-spectrum of antibodies. Fractionation of IgG subclasses can also be performed using recombinant protein A.

Because of its high affinity for immunoglobulins of many species and its low level of non-specific binding, recombinant protein A is useful for the study of cell surface antigens and receptors, and for detecting antibody-secreting cells.

Recombinant protein A is produced in an *E. coli* strain which contains the gene for mature protein A from *Staphylococcus aureus* Cowan I strain[1]. Recombinant protein A contains four IgG binding domains labelled D, A,B,C and repeating C-terminal regions labelled S-1 through S-11. These regions are believed to be involved in binding protein A to the cell wall of *Staphylococcus aureus*. No amino acids from the protein A signal sequence are included in the recombinant construction.

Recombinant protein A is a salt-free, lyophilized powder. Protein is determined using biuret methodology.

**Molecular Weight:** The molecular weight of recombinant protein A is 45,000 as determined by SDS-PAGE. This is similar to the 45,216 molecular weight calculated from the gene sequence.

**Purity:** By SDS-PAGE gel electrophoresis, the product shows a single band of approximately 45 kDa.

**Activity:** The specific activity of recombinant protein A is equal to the most active protein A preparations extracted from *Staphylococcus aureus*. The disassociation constants for both *Staph. aureus* derived and recombinant protein A to rabbit IgG are $1x10^{-9}$M at pH 7.4, as measured by a solid phase competition RIA.

Protein A and IgG combine to form a precipitation complex. In an assay containing 0.5 mg/ml protein A and varying concentrations of IgG, recombinant protein A will precipitate 3.5 moles of IgG per mole. Protein A derived from *Staph. aureus* precipitates 1.6 moles IgG per mole in this assay. Protein A from both sources reveal a $K_{0.5}$ of 0.15 for the precipitation reaction with a Hill coefficient of 1.75.

Solid-phase ELISA assays show recombinant protein A to have equal or greater binding capacity than several commercial protein A preparations, when used as a "bridge" between purified rabbit IgG and peroxidase conjugated protein A.

**IgG Binding:** ~99%

**Reference:**

– Colbert, D. et al., **J. Biological Response Modifiers 3**, 255, 1984.

**Note:** This product may contain a preservative such as sodium azide, thimerosal or proclin. Please see lot specific chemical credential for preservative information.