

ProteinIso® Protein G Resin

Cat. No. DP401

Version No. Version 2.0

Storage: at 2-8°C (20% ethanol) for two years

Description

ProteinIso® Protein G Resin is an affinity chromatography resin with high binding capacity for IgG. The recombinant protein G ligand is coupled to highly cross-linked agarose. *ProteinIso*® Protein G Resin is suitable for purification of monoclonal antibody, polyclonal antibody.

Resin Specifications

| Resin | Cross-linked 4% agarose | |
|--|----------------------------|--|
| Ligand | r-Protein G | |
| Shape | Sphere | |
| Pore size | 90 μm (45-165) | |
| Support density | 3 mg Protein G/ml wet gel | |
| Binding capacity | 20-25 mg h-IgG /ml wet gel | |
| Maximum flow rate (25°C) | 300 cm/h | |
| Recommended flow rate | <150 cm/h | |
| Highest resistance of atmospheric pressure | 0.3 Mpa | |
| pH stability | 3~10 | |

Procedures

- 1. Prepare protein G purification column
- (1) Thoroughly resuspend the protein G resin to achieve a homogeneous suspension of the resin in the 20% ethanol storage buffer.
- (2) Immediately transfer the resin into a purification column. Ensure that the bottom of the column is plugged with a stopper. Close the valve of the column. Allow the resin to settle.
- (3) Equilibrate the column with 5~10 bed volume of equilibration buffer.
- 2. Prepare samples

To avoid blocking column, samples should be centrifuged and filtrated with 0.45 μm filter before loading.

- 3. Load samples and wash
 - Load samples and wash with 5~10 bed volume of equilibration buffer and collect the flow-through in one tube.
- 4. Elute
 - Elute antibodies with elution buffer.
 - Collect the elution containing the target immunoglobulin and immediately neutralize to pH>7.0 with neutralization buffer. The elution conditions are closely related with binding strength and stability of antibody. When necessary, optimize the elution buffer.
- 5. Regeneration of Protein G Resin
- (1) Wash the column/resin with $3\sim5$ bed volume of 0.1 M citric acid or 0.1 M citric acid /20% ethanol and then 5 bed volume of PBS buffer (pH=7.0).

Or

- (2) 3~5 bed volume of 0.05 M NaOH/1 M NaCl or 6 M GuHCl, and then 5 bed volume of deionized water.
- (3) Store column/resin in 20% ethanol.





Notes

- Samples should be centrifuged and filtrated with 0.45 µm filter before loading.
- Equilibration Buffer

20 mM PB, 150 mM KCl pH 7.0

• Elution Buffer

20 mM citric acid pH 3.0-4.0; or 100 mM glycine pH 3.0; or 20 mM sodium acetate pH 3.0-4.0.

- Neutralization Buffer
- 1 M Tris-HCl pH 9.0.

Affinity of Protein A/G for IgG Types

Agarose affinity medium immobilized with Protein A and G can both be used in antibody purification.

The affinity of Protein A and protein G for immunoglobulins varies with different sources and subclasses. The following table compares the IgG binding capacities of protein A and G for reference.

It should be noted that the strength of antibody binding ability does not directly reflect the quality of antibody purification effect.

| Sources | IgG Subtype | Affinity for Protein A | Affinity for Protein G |
|---------|-------------|------------------------|------------------------|
| Human | IgG1 | ++++ | ++++ |
| | IgG2 | ++++ | ++++ |
| | IgG3 | - | ++++ |
| | IgG4 | ++++ | ++++ |
| Mouse | IgG1 | + | ++++ |
| | IgG2a | ++++ | ++++ |
| | IgG2b | +++ | +++ |
| | IgG3 | ++ | +++ |
| Rabbit | IgG | ++++ | +++ |
| Goat | IgG | - | ++ |
| Horse | IgG | ++ | ++++ |
| Gog | IgG | ++ | + |
| Bovine | IgG | ++ | ++++ |
| Porcine | IgG | +++ | +++ |
| Monkey | IgG | ++++ | ++++ |

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