

# **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  $5 \sim 10$  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
- Wash the column/resin with 3~5 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

- $\bullet$  Samples should be centrifuged and filtrated with 0.45  $\mu m$  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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