

## ProteinIso<sup>®</sup> GST Resin

Please read the manual carefully before use

**Cat. No.** DP201

**Version No.** Version 1.1

**Storage:** at 2°C-8°C for two years

### Description

ProteinIso<sup>®</sup> GST Resin is an affinity chromatography medium that uses glutathione as the ligand and agarose as the matrix. It can efficiently and specifically adsorb GST-tagged fusion proteins, and allows elution under mild, non-denaturing conditions using reduced glutathione. This process preserves the antigenic activity and function of the proteins throughout purification. It is suitable for purifying GST-tagged proteins, glutathione S-transferase, or glutathione-dependent proteins.

### Resin Specifications

Resin	Cross-linked 4% agarose
Ligand	Glutathione
Shape	Sphere
Pore size	90 μm
Ligand density	8 mg GSH/ml wet gel
Binding capacity	10~12 mg GST-fusion protein/ml wet gel (MW 42 kDa) 3.5 mg GST/ml wet gel (rat liver)
Maximum flow rate (25°C)	450 cm/h
Recommended flow rate	<150 cm/h
Highest resistance of atmospheric pressure	0.3 Mpa
pH stability	3~10
Chemical stability	Stable in common aqueous buffers, 70% ethanol, 30% isopropanol, 6 M guanidine hydrochloride, and 1 M acetic acid (pH 4.0)
Physical stability	Variations in the solution's pH or ionic strength result in less than a 2% change in the volumetric change rate of the resin

### Procedures

The procedures typically include: column packing, equilibration, sample loading, washing, elution, and regeneration.

1. Column packing: Resuspend the resin and add an appropriate amount to the chromatography column based on the quantity of protein to be purified. Allow the resin to settle.
2. Equilibration: Equilibrate the resin with 5-10 CV of equilibration buffer until the conductivity and pH of the flow-through remain constant (matching those of the equilibration buffer).
3. Sample loading: The sample buffer should be as consistent as possible with the equilibration buffer. To prevent column clogging, the sample should be centrifuged or filtered through a 0.45 μm filter prior to loading.
4. Washing: After sample loading, wash the resin with 5-10 CV of equilibration buffer and collect the flow-through.
5. Elution: Elute with elution buffer (50 mM Tris-HCl pH 8.0, 10 mM reduced glutathione). The concentration of glutathione should be adjusted accordingly based on the binding affinity of the target protein. Glutathione is prone to oxidation and should be prepared fresh immediately before use. Collect the flow-through.
6. Regeneration: After several uses (the number of uses depends on the type and source of the raw material and the experimental requirements), precipitates, denatured proteins, and non-specifically adsorbed proteins may accumulate on the resin. This may lead to a decrease in the binding capacity of the resin, and regeneration is required.



- (1) To remove precipitates and denatured proteins, wash the resin with 2 CV of 6 M GuHCl, then immediately equilibrated to neutrality with 5 CV of neutral PBS buffer.
  - (2) To remove hydrophobic binding proteins, lipoproteins, and lipidic substances, wash the resin with 3-4 CV of 70% ethanol or 30% isopropanol (for more than 20 minutes), followed by washing with 3-5 CV of pure water.
  - (3) Or wash the resin with 2 CV of 0.01-0.1 M NaOH, then immediately equilibrated to neutrality with 5 CV of neutral PBS buffer.
- (It is recommended to prioritize washing methods (1) or (2).)

#### Notes

- To prevent column clogging, the sample should be filtered through a 0.45  $\mu\text{m}$  filter prior to loading.
- After repeated use, the binding capacity of the resin may change, which is dependent on the specific sample being purified. To avoid cross-contamination, do not use the same resin to purify different proteins.
- Recommended formulation for equilibration buffer  
140 mM NaCl, 2.7 mM KCl, 10 mM  $\text{Na}_2\text{HPO}_4$ , 1.8 mM  $\text{KH}_2\text{PO}_4$ , pH 7.3
- Recommended formulation for elution buffer  
50 mM Tris-HCl pH 8.0, 10 mM reduced glutathione. (The concentration of glutathione should be adjusted accordingly based on the binding affinity of the target protein. Glutathione is prone to oxidation and should be prepared fresh immediately before use.)

**For research use only, not for clinical diagnosis.**

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