Technical literature is available at: www.mesgenbio.com. E-mail MesGen Technical Services if you have questions on use of this system: tech@mesgenbio.com



Description

MesGen Glutathione Resin (MPB90) is an affinity chromatography medium designed for easy, one-step purification of recombinant glutathione S-transferase (GST) fusion proteins and other glutathione binding proteins expressed in *E. coli*, insect cells and mammalian cells. The recombinant GST fusion proteins can be purified directly from pre-treated cell lysate using Glutathione Resin. It is the excellent choice for high performance purifications.

Binding Capacity

≥20 mg of purified recombinant glutathione S-transferase (GST) per milliliter of settled resin

Average particle size

60 µm

Purification Buffers

- Equilibration/Wash Buffer: 50mM Tris, 150mM NaCl, pH 8.0. Note: Suggest adding protease inhibitors, such as Protease Inhibitor Cocktail (Product No. MG2284) or PMSF (Product No. MG0754) to protect proteins from degradation.
- 2. Elution Buffer: 50mM Tris, 150mM NaCl, pH 8.0 containing 10mM reduced glutathione.

Procedure for Purification of GST-tagged Proteins using a Gravity-flow Column

Note: Perform the procedure at room temperature or at 4°C.

- 1. Pack column with an appropriate amount of Glutathione Resin (1ml settled resin could bind ≥20 mg of purified recombinant GST).
- 2. For best results, prepare sample by mixing protein extract with Equilibration/Wash Buffer so the total volume equals at least two resin-bed volumes. Other ratios may be used but need to be determined empirically.
- 3. Equilibrate column with 10 resin-bed volumes of Equilibration/Wash Buffer. Using a flow rate of 0.5ml/minute, allow buffer to drain from resin.
- 4. Add the prepared protein extract to the resin.
- 5. Wash resin with 10-20 resin-bed volumes of Equilibration/Wash Buffer.
- 6. Elute GST-tagged protein from the resin with two resin-bed volumes of Elution Buffer. Repeat this step twice, collecting each fraction in a separate tube.
- 7. Monitor protein elution by measuring the absorbance of the fractions at 280nm or by Protein Assay Dye Reagent (Product No. MG1030). The eluted protein can be directly analyzed by SDS-PAGE.

Procedure for Glutathione Resin Regeneration

The Glutathione Agarose may be used at least five times without affecting protein yield or purity. Between each use, perform the procedure described below to remove residual glutathione and any nonspecifically adsorbed protein. To prevent cross-contamination of samples, designate a given column to one specific fusion protein.

- 1. Apply 2 resin-bed volumes of Buffer (0.1 M Tris HCl + 0.5 M NaCl, pH 8.5).
- 2. Apply 2 resin-bed volumes of Buffer (0.1 M natrium aceticum + 0.5 M NaCl, pH 4.5)
- 3. Apply 3 resin-bed volumes of PBS.
- 4. Apply 3 resin-bed volumes of ultrapure water.
- 5. Wash the column with 5mL of 0.05% sodium azide (in water). Cap bottom and top of column. Store at 4°C.

Storage condition

2-8°C

Shelf life

18 months when stored unopened

For Research Use Only. Not for use in diagnostic procedures.