GST protein purification beads

DESCRIPTION

GST fusion protein purification magnetic beads are a new type of functional material designed for efficient and rapid purification of glutathione sulfhydryl transferase (GST) fusion proteins. It can be purified directly from biological samples in one step by magnetic separation. The target protein, which greatly simplifies the purification process and improves the purification efficiency, is suitable for the convenient purification of GST fusion protein in scientific research and industrial fields.

PRODUCT INFORMATION

Type	GST protein purification beads
Size	30 ~ 50 μm
GSH ligand content	20 ~ 30 μmol/mL
Fusion protein binding capacity	10 mg/mL
Suspended concentration	10%（v/v）Magnetic bead suspension
Buffer	20%（v/v）Ethanol
Storage	2~8℃

OPERATING PROCEDURES

1. Buffer solution preparation

1.1 Buffer A: 140 mM NaCl, 2.7 mM KCl, 10 mM Na2HPO4, 1.8 mM KH2PO4, pH 7.4.

1.2 Buffer B: 50 mM Tris-HCl, 10 mM reduced glutathione, pH 8.0 Preparation method:

0.1 M Tris solution 50 mL, 0.307 g reduced glutathione, then adjust the pH to 8.0 with 0.1

M hydrochloric acid, Add deionized water to 100 mL.

2. Sample processing

2.1 Protein expression in E. coli, yeast and other cells: the expressed cells are diluted with an appropriate amount of Buffer A, and protease inhibitors are added; the cells are lysed by ultrasonic in an ice bath, which is the crude protein sample.

2.2 Extracellular expression protein: Take the extracellular expression supernatant and dilute it with the same amount of Buffer A to obtain a crude protein sample.

2.3 Intracellular expression of animal cells: Take an appropriate amount of animal cells, wash with an appropriate amount of PBS, and discard the supernatant; use an appropriate amount of 1% (v/v) Triton X-100 or 1% (v/v) NP-40 Resuspend in Buffer A; add protease inhibitors (such as PMSF with a final concentration of 1 mM); place on ice for 10 minutes to obtain a crude protein sample.

3. Pretreatment of magnetic beads

3.1 Put the magnetic bead product on the vortex mixer and mix well, use a pipette to take 10 mL of the magnetic bead suspension into the centrifuge tube; 3.2 Place the centrifuge tube on the magnetic separator, and after the solution becomes clear, pipette Go to the supernatant;

3.3 Add 5~10 mL Buffer A to the above centrifuge tube containing magnetic beads, close the lid, and vortex for 15 seconds to resuspend the magnetic beads. Place the centrifuge tube on the magnetic separator, magnetically separate, remove the supernatant, and repeat the washing twice.

4. The target protein is bound to the magnetic beads

4.1 Use 10 mL Buffer A to suspend 1 g wet weight of the bacterial cells, break and lyse, and then it will be a crude protein sample;

4.2 Add the crude protein sample to the centrifuge tube containing the pre-treated magnetic beads, and close the centrifuge tube cap tightly;

4.3 Place the centrifuge tube in a vortex mixer and shake for 15 seconds, then place it on a rotary mixer, and rotate and mix at room temperature for 20-30 minutes;

4.4 Place the centrifuge tube on the magnetic separator for magnetic separation, and

transfer the supernatant to a new centrifuge tube for subsequent testing. Remove the centrifuge tube from the magnetic separator for subsequent washing steps.

5. Magnetic bead washing

5.1 Add 5~10 mL of Buffer A to the centrifuge tube containing magnetic beads, spin and mix for 2 minutes, magnetically separate, and transfer the cleaning solution to a new centrifuge tube for sampling and testing;

5.2 Add 5~10 mL of Buffer A to the centrifuge tube containing magnetic beads, resuspend the magnetic beads, and transfer the magnetic bead suspension to a new centrifuge tube to avoid non-specific adsorption proteins on the original centrifuge tube wall from contaminating the target protein; Separate and transfer the supernatant to the cleaning solution collection tube.

6. Elution of target protein

Add 2~5 mL Buffer B to the centrifuge tube, cap the centrifuge tube tightly, then place the centrifuge tube on the rotary mixer, rotate and mix at room temperature for 2 minutes; magnetically separate, collect the eluate into a new centrifuge tube, that is Is a purified target protein sample.

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