

Glutathione Magnetic Agarose Beads

1 Contents

Product Name	HY-K0234-1 mL	HY-K0234-5 mL	HY-K0234-25 mL
Glutathione Magnetic Agarose Beads (Settled Beads)	1 mL	5 mL	5 mL× 5

2 General Information

MCE Glutathione Magnetic Agarose Beads are based on magnetic cross-linked agarose, effectively purify high levels of overexpressed GST-tagged fusion proteins at a variety of scales. MCE Glutathione Magnetic Agarose Beads have high protein-binding capacity and stability, making it ideal for high performance purification of GST-tagged fusion proteins expressed in E. coli, yeast, insect and mammalian expression systems.

Glutathione Magnetic Agarose Beads are removed from the solution manually by using a magnetic stand or automatically by using an instrument. With high loading of GST-tagged protein and high specificity, Glutathione Magnetic Agarose Beads are also suitable for GST Pull-down.

3 Characteristics

Bead Diameter	30-100 μm
Binding Capacity	5-10 mg GST protein/mL Settled Beads
Application	Pull-down, Protein Purification
Storage Solution	20% slurry in a 20% ethanol solution

4 General Protocol

Two protocols are recommended according to further usage.

4.1 GST-tagged Protein Purification

Recommended Buffer

Binding/Wash Buffer	PBS: 140 mM NaCl, 2.7 mM KCl, 10 mM Na_2HPO_4 , 1.8 mM KH_2PO_4 , pH 7.4
Elution Buffer	50 mM Tris-HCl, 10-20 mM GSH, pH 8.0

(1) Preparation of Magnetic Agarose Beads

a. Resuspend the Glutathione Magnetic Agarose Beads in the vial (tilt and

rotate for 2 minutes or gently pipette for 10 times, do not vortex). Transfer appropriate amount of Glutathione Magnetic Agarose Beads into a new tube.

b. Add appropriate amount of Binding/Wash Buffer to the beads and gently pipette to mix. Place the tube into a magnetic stand to collect the beads against the side of the tube. Remove and discard the supernatant. Repeat this step for 2 times.

(2) Protein Binding

a. Add appropriate amount of cell lysate (the sample containing GST-tagged protein) to the washed beads and incubate for 30 minutes at room temperature or 4°C at 1 hour while gently rotating the tube.

b. Place the tube into a magnetic stand to collect the beads against the side of the tube. The supernatant was collected in a new centrifuge tube for subsequent detection.

(3) Washing

Add 2× beads-volumes of Binding/Wash Buffer to the beads complex and mix gently. Place the tube into a magnetic stand to collect the beads against the side of the tube. Remove and discard the supernatant. Repeat this step for 2 times.

(4) Elution

Add 3-5× beads-volumes of Elution Buffer to each tube. Incubate with gentle shaking or on a rotator for 5-10 minutes at room temperature. Place the tube into a magnetic stand to collect the beads and transfer the supernatant to a new tube.

4.2 GST Pull-Down

Recommended Buffer

Binding/Wash Buffer	PBS: 140 mM NaCl, 2.7 mM KCl, 10 mM Na_2HPO_4 , 1.8 mM KH_2PO_4 , pH 7.4
Elution Buffer	50 mM Tris-HCl, 10-20 mM reduced glutathione, pH 8.0

(1) Preparation of Magnetic Agarose Beads

- a. Resuspend the Magnetic Agarose Beads in the vial (tilt and rotate for 2 minutes or gently pipette for 10 times, do not vortex). Transfer 100 μ L of Glutathione Magnetic Agarose Beads into a new tube.
- b. Add 500 μ L Binding/Wash Buffer to the beads and gently pipette to mix. Place the tube into a magnetic stand to collect the beads against the side of the tube. Remove and discard the supernatant. Repeat this step for 2 times.

(2) Binding of GST-tagged Protein

- a. Add 500 μ L of the sample containing GST-tagged protein to the washed beads and incubate for 30 minutes at room temperature or 4°C at 1 hour while gently rotating the tube. Place the tube into a magnetic stand to collect the beads against the side of the tube. Remove and discard the supernatant.
- b. Add 1 mL Binding/Wash Buffer to the beads complex and mix gently. Place the tube into a magnetic stand to collect the beads against the side of the tube. Remove and discard the supernatant. Repeat this step for 2 times.

(3) Binding of Target Protein

- a. Add 500 μ L of the target protein to the tube and incubate for 30 minutes at room temperature while gently rotating the tube. Place the tube into a magnetic stand to collect the beads against the side of the tube. Remove and discard the supernatant.
- b. Add 1 mL Binding/Wash Buffer to the tube and mix gently. Place the tube into a magnetic stand to collect the beads against the side of the tube. Remove and discard the supernatant. Repeat this step for 2 times.

(4) Elution

Add 3-5 \times beads-volumes of Elution Buffer to each tube. Incubate with gentle shaking or on a rotator for 5-10 minutes at room temperature. Place the tube into a magnetic stand to collect the beads and transfer the supernatant to a new tube.

5 Storage

Store at 2-8°C, and is stable for up to 2 years.

6 Precautions

- (1) Do not centrifuge, dry or freeze the magnetic agarose beads.
- (2) To minimize protein degradation, protease inhibitor cocktails (MCE Cat. No.: HY-K0010, HY-K0011) are highly recommended.
- (3) For the best experimental performance, it is recommended to use the MCE magnetic stand (Cat. No.: HY-K0200).
- (4) If the target protein is not completely eluted, it may be due to the oxidation of reduced glutathione in Elution Buffer. So it is recommended to use a freshly prepared Elution Buffer.
- (5) Adding 1-10 mM DTT to samples and buffers can improve the binding of GST fusion proteins to magnetic agarose beads.
- (6) This product is for R&D use only, not for drug, house hold, or other uses.