

Anti-c-Myc Magnetic Beads

1 Contents

Cat. No.	Product Name	Package
HY-K0206-1 mL	Anti-c-Myc Magnetic Beads	1 mL
HY-K0206-5 mL	Anti-c-Myc Magnetic Beads	1 mL × 5

2 General Information

MCE Anti-c-Myc magnetic beads are used for immunoprecipitation (IP) of specific c-Myc-tagged proteins expressed in bacterial and mammalian cells and in vitro expression systems. Anti-c-Myc magnetic beads are based on amino magnetic beads, with 2 μm particle size, covalently coupling with high quality mouse IgG₁ monoclonal antibody that recognizes the c-Myc-epitope tag (EQKLISEEDL) derived from the human c-Myc protein. Magnetic beads are removed from the solution manually by using a magnetic stand or automatically by using an instrument. With high loading of c-Myc-tagged protein and high specificity, Anti-c-Myc Magnetic Beads are also suitable for Co-immunoprecipitation and purification of c-Myc-tagged protein.

3 Characteristics

Composition	Mouse IgG ₁ monoclonal antibody covalently coupled to a blocked magnetic bead surface
Bead Diameter	2 μm
Binding Capacity	>1 mg protein/mL of beads
Application	IP, Protein Purification
Recommended Dose	10-20 μL for per 500 μL cell lysates

4 General Protocol

Recommended Wash Buffer

PBST: 1×PBS + 0.5% Triton X-100, pH 7.4

1. Preparation of Magnetic Beads

1.1 Resuspend the Magnetic Beads in the vial (tilt and rotate for 2 minutes or gently pipette for 10 times, do not vortex). Transfer 20 μL of Anti-c-Myc Magnetic Beads suspension into a new tube.

1.2 Add 500 μL of wash buffer to the beads and gently pipette to mix. Place the tube into a magnetic stand (MCE Cat. No.: HY-K0200) to collect the beads against the side of the tube. Remove and discard the supernatant. Repeat this step for 2 times.

2. Protein Binding

2.1 Add 500 μL of cell lysate (the sample containing c-Myc-tagged protein) to the washed beads. For Ag binding, incubate for 2 hours at room temperature or overnight at 4°C while gently rotating the tube.

2.2 Place the tube into a magnetic stand to collect the beads against the side of the tube. Remove and discard the supernatant.

Note: Occasional aggregation of magnetic beads during the binding process doesn't affect experimental results.

3. Washing

Add 500 μL of wash buffer to the Magbeads-Ag 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 4 times.

4. Elution & Detection

Add 50 μL of 1× SDS-PAGE loading buffer into the tube with Magbeads-Ag complex and boil for 5 minutes. Cool and place the tube into a magnetic stand to collect the beads against the side of the tube. Keep the supernatant containing the target antigen for SDS-PAGE analysis.

5 Storage

Store at 4°C, and is stable for up to 2 years.

6 Precautions

- 1 The pH of Anti-c-Myc Magnetic Beads is 6-8.
- 2 Do not centrifuge, dry or freeze the magnetic beads. Centrifuging, drying or freezing will cause the beads to aggregate and lose binding affinity.
- 3 For best results, determine optimal conditions for expression of c-Myc-tagged fusion protein before attempting immunoprecipitation.
- 4 To minimize protein degradation, protease inhibitor cocktails (MCE Cat. No.: HY-K0010, HY-K0011) are highly recommended.
- 5 For the best experimental performance, it is recommended to use the MCE magnetic stand (Cat. No.: HY-K0200).
- 6 Do not use cell lysate containing dithiothreitol (DTT). DTT may cause the c-Myc antibody to leach from the beads.
7. This product is for R&D use only, not for drug, house hold, or other uses.

Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

7 Troubleshooting

Problem	Possible Cause	Solution
High background	Nonspecifically binding of protein to the antibody, magnetic beads or EP tubes	Pre-clear lysate to remove nonspecific binding proteins
		After suspending beads for the final wash, transfer the entire sample to a clear EP tube and then magnetic separation or centrifugation
	Washing times were not sufficient	Increase the number and time of washes
Little or no c-Myc-tagged protein is detected	No or minimal tagged protein was expressed	Verify protein expression by SDS-PAGE or Western blot analysis of the lysate using an c-Myc-tagged positive control as a reference
		Increase the amount of lysate used for IP
	Tagged protein degraded	Prepare fresh lysate
		Use appropriate protease inhibitors (MCE Cat. No.: HY-K0010, HY-K0011)
	Incubation time was inadequate	Prolong the incubation time
	Interfering substance was contained	Do not use cell lysate containing dithiothreitol (DTT), 2-mercaptoethanol, or other reducing agents
Excessive detergent concentration may interfere with the antibody-antigen interaction		