

## V5 Tag Immunomagnetic Beads

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### Description

TargetMol's V5 Tag Immunomagnetic Beads can specifically bind to V5-tagged proteins and can be utilized for immunoprecipitation (IP) of proteins, protein complexes, protein-nucleic acid complexes, and other antigens. This product is suitable for antigen samples derived from cell lysates, cell culture supernatants, serum, ascites, and other sources.

### Features

- Low non-specific adsorption
- High efficiency, high yield, low consumption
- Flexible and simple operation
- Reliable experimental results with high reproducibility

### Product Information

Property	Characteristics
Matrix	Silica-based magnetic beads
Particle Size	200 nm
Bead Concentration	10 mg/mL
Binding Capacity	≥0.6 mg V5-tagged protein / mL magnetic beads
Ligand	Mouse anti-V5 monoclonal antibody
Recommended Applications	IP, Co-IP

### Instructions

#### Reagents to be Prepared by the User

Reagent	Optional Formulation
Washing Buffer (1×)	TBST: 50 mM Tris-HCl, 150 mM NaCl, 0.1% (v/v) Tween-20, pH 7.4
Acid Elution Buffer	0.1 M Glycine, 0.1% (v/v) Tween-20, pH 2.5
Neutralization Buffer	1 M Tris-HCl, pH 9.0

#### 1. Cell Lysate Preparation

Treat cell samples with an appropriate lysis buffer, prepare the cell lysate according to standard protocols, and place on ice for immediate use, or store at -20°C for long-term storage.

## 2. Magnetic Bead Pretreatment

- 1) Resuspend the immunomagnetic beads thoroughly by vortexing for 1 min. Transfer 25–50  $\mu\text{L}$  of the bead suspension into a 1.5 mL EP tube.
- 2) Add 500  $\mu\text{L}$  of Washing Buffer to the EP tube, and gently invert the tube several times to resuspend the beads. Place the EP tube into a magnetic separator and leave it undisturbed for 1 min to achieve magnetic separation. Aspirate and discard the supernatant, then remove the EP tube from the separator.
- 3) Repeat the above washing steps twice.

## 3. Immunoprecipitation Protocol

- 1) Add 500  $\mu\text{L}$  of the prepared cell lysate to the EP tube containing the pretreated beads. Place the tube on a rotary mixer and incubate with rotation at 37°C for 30 min. If the binding affinity is weak, incubation can be performed at room temperature for 1 h, or at 4°C overnight.
- 2) Following incubation, perform magnetic separation. Discard or save the supernatant for subsequent analysis.
- 3) Add 500  $\mu\text{L}$  of Washing Buffer to the EP tube to wash the beads. Perform magnetic separation, aspirate and discard the supernatant, and remove the EP tube. Repeat the bead washing steps 3 times.

## 4. Antigen Elution

Several antigen elution protocols are provided below. Users may select the appropriate elution method based on the requirements of downstream detection applications.

- Denaturing Elution Method: Samples eluted by this method are suitable for SDS-PAGE analysis. Add 100  $\mu\text{L}$  of SDS-PAGE Loading Buffer (C0190) to the EP tube, mix thoroughly, and heat at 95°C for 5 min. Then, perform magnetic separation or centrifugation (room temperature, 13,000  $\times$  g, 10 min), collect the supernatant, and proceed with SDS-PAGE analysis.
- Acid Elution Method: Add 100  $\mu\text{L}$  of Acid Elution Buffer to the EP tube, and incubate on a rotary mixer at 37°C for 5–10 min. Subsequently, perform magnetic separation or centrifugation, and collect the supernatant. If neutralization of the acidic eluate is required, add 50  $\mu\text{L}$  of Neutralization Buffer to 100  $\mu\text{L}$  of the collected eluate to adjust the pH to neutral.

## Storage Conditions

Store at 4°C for 2 years.




## Precautions

1. Avoid freezing the magnetic beads. Beads must be maintained in the storage solution to prevent drying out.
2. To minimize the loss of magnetic beads, the duration for each magnetic separation step should not be less than 1 min.
3. Before withdrawing beads from the stock tube, vortex thoroughly to ensure a homogenous suspension. Take care to avoid generating air bubbles during operation.
4. It is recommended to use high-quality pipette tips and reaction tubes to prevent material loss caused by the adhesion of beads and solutions to plastic surfaces.
5. Operators may analyze the binding efficiency of the antibody and antigen to the magnetic beads by testing the supernatants collected during the antibody-binding and antigen-binding steps based on experimental needs.

6. In IP experiments, the binding affinity varies among different types of antibodies and antigens. If the buffer system recommended in this manual fails to yield optimal experimental results, operators may screen and formulate customized buffers for optimization.
7. This product is intended solely for scientific research use by professional personnel. It must not be used for clinical diagnosis or treatment, food or drugs, and must not be stored in residential areas.
8. For your safety and health, please wear a lab coat and disposable gloves during operation.

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