

Anti DYKDDDDK Antibody-HRP

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1. Product Description

1.1 Product Introduction

The DYKDDDDK peptide is one of the most commonly used tags in fusion protein expression and purification. The DYKDDDDK peptide is also often referred to as the Flag® peptide, which is a registered trademark of Sigma. It can be fused to the N-terminus, C-terminus, or within the sequence of a fusion protein. Highly hydrophilic, it is more readily displayed on the surface of the fusion protein, minimizing the likelihood of interfering with the protein's function. This tag offers an additional advantage: when fused to the N-terminus of the target protein, it can be cleaved by Enterokinase (EK protease) at the C-terminal side of its aspartic acid residue, leaving no residual amino acids. Antibodies against the DYKDDDDK tag enable precise detection and purification of DYKDDDDK-tagged fusion proteins. By conjugating Horseradish Peroxidase (HRP) to the anti-DYKDDDDK antibody through a specific process, the tagged fusion protein can be rapidly detected in Western blot (WB) assays. Upon adding chemiluminescent substrates A and B (typically luminol and hydrogen peroxide), catalyzed by HRP, luminol reacts with hydrogen peroxide to form an unstable peroxide intermediate. This intermediate decomposes, generating an electronically excited state species. When this species returns to the ground state, light is emitted, enabling rapid detection of the DYKDDDDK-tagged fusion protein.

1.2 Basic Information

Antibody Source: Recombinant Antibody

Cross Reactivity: DYKDDDDK-tagged Fusion Proteins

Clonality: Monoclonal Antibody

Conjugate: Horseradish Peroxidase (HRP)

Purification Method: Protein A Affinity Purified

Storage Buffer: 1× PBS (pH 7.4), 0.05% proclin300, 50% Glycerol

Storage Conditions: Shipped on dry ice. Store at -20°C for up to 2 years. Avoid repeated freeze-thaw cycles..

2. Product Applications

WB=1:30000-1:100000

ELISA=1:5000-1:10000.

3. Additional information

3.1 This product is an HRP-conjugated antibody and must be stored protected from light at -20°C or -80°C.

3.2 This product is highly sensitive. For Western blot (WB), please follow the recommended dilution ratios provided in the manual to avoid non-specific signals.

3.3 NC or PVDF membranes must be activated with methanol prior to transfer. After transfer, ensure thorough coverage with antibody dilution buffer, and avoid creating bubbles during both the transfer and antibody incubation steps.

3.4 This product can generate strong signals after 30-60 minutes of incubation at 37°C. If incubating at room temperature, prolong the duration appropriately.

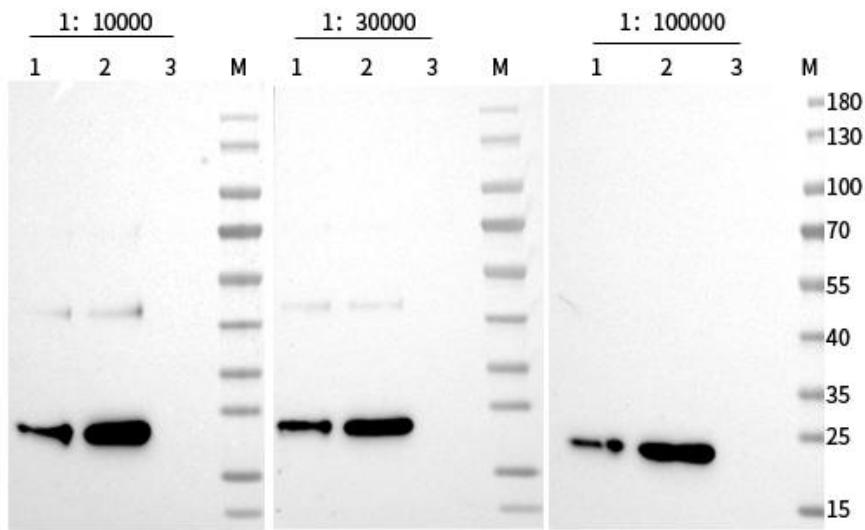
4. Reference Information

1) Expression System: Secretory expression in 293 cells.

2) Protein Information: Recombinant protein with a DYKDDDDK tag, approximate size ≈25 kDa.

3) Loading Order: Lane 1: 10 ng of purified target protein; Lane 2: 50 ng of purified target protein; Lane 3: Bovine Serum Albumin (BSA).

- 4) Antibody Diluent Volume: 10 mL.
- 5) Antibody Incubation Conditions: 37°C with shaking for 30 minutes.
- 6) Detection: ECL chemiluminescent substrate.
- 7) Results:



1: Purified Target Protein (10 ng); 2: Purified Target Protein (50 ng); 3: (BSA)

8) Conclusion: When the Anti DYKDDDDK Antibody-HRP was diluted at ratios of 1:10,000, 1:30,000, and 1:100,000 (in a total antibody diluent volume of 10 mL) and incubated with shaking at 37°C for 30 minutes, distinct signals were observed. The target band appeared at approximately 25 kDa, with weaker aggregate bands visible around 50-55 kDa. The signal intensity decreased progressively as the dilution ratio increased.

5. Related Products

Product	Cat.No.	Size
Anti DYKDDDDK Antibody-HRP	BP5013-01	20µl
	BP5013-02	100µl
	BP5013-03	1ml