

#12698 Store at -20°C

## His-Tag (D3I1O) XP® Rabbit mAb



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Applications:	Reactivity:	Sensitivity:	Source/Isotype:
WB, W-S, IP, IF-IC, FC-FP, ChIP	All	Transfected Only	Rabbit IgG

### Product Usage Information

For optimal ChIP results, use 5 µl of antibody and 10 µg of chromatin (approximately 4 x 10<sup>6</sup> cells) per IP. This antibody has been validated using SimpleChIP® Enzymatic Chromatin IP Kits.

Application	Dilution
Western Blotting	1:1000
Simple Western™	1:50 - 1:250
Immunoprecipitation	1:50
Immunofluorescence (Immunocytochemistry)	1:400
Flow Cytometry (Fixed/Permeabilized)	1:800
Chromatin IP	1:100

### Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

### Specificity / Sensitivity

His-Tag (D3I1O) XP® Rabbit mAb recognizes recombinant proteins containing the 6xHis epitope tag. The antibody recognizes the 6xHis-tag fused to either the amino or carboxy terminus of targeted proteins in transfected cells.

### Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues of the 6xHis epitope tag.

### Background

Epitope tags are useful for the labeling and detection of proteins using immunoblotting, immunoprecipitation, and immunostaining techniques. Because of their small size, they are unlikely to affect the tagged protein's biochemical properties.

A variety of plasmids contain DNA that encodes an amino-terminal tag consisting of six histidine (6xHis) residues followed by an extended multiple cloning site. The 6xHis tag on the expressed recombinant proteins allows for efficient coupling to Ni<sup>2+</sup> affinity resins and purification by single step chromatography (1).

As is the case with other protein tag systems (2), this polyhistidine tag can often be cleaved at sites recognized by proteases such as thrombin and enterokinases to isolate the protein of interest (1).

### Background References

1. Kroll, D.J. et al. (1993) *DNA Cell Biol* 12, 441-53.
2. di Guan, C. et al. (1988) *Gene* 67, 21-30.

### Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

### Western Blot Buffer

**IMPORTANT:** For western blots, incubate membrane with diluted primary antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

### Applications Key

**WB:** Western Blotting **W-S:** Simple Western™ **IP:** Immunoprecipitation  
**IF-IC:** Immunofluorescence (Immunocytochemistry) **FC-FP:** Flow Cytometry (Fixed/Permeabilized)  
**ChIP:** Chromatin IP

### Cross-Reactivity Key

**H:** human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **Mi:** mink **C:** chicken **Dm:** D. melanogaster  
**X:** Xenopus **Z:** zebrafish **B:** bovine **Dg:** dog **Pg:** pig **Sc:** S. cerevisiae **Ce:** C. elegans **Hr:** horse  
**GP:** Guinea Pig **Rab:** rabbit **All:** all species expected

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