

Datasheet for 200-301-382S**6X His Epitope Tag Antibody****Overview**

Description:	Anti-6X His Epitope Tag (MOUSE) Monoclonal Antibody - 200-301-382S
Item No.:	200-301-382S
Size:	25 µL
Applications:	ELISA, WB
Reactivity:	His-Tag
Host Species:	Mouse

Product Details

Background:	6X His antibody is an Epitope tag which is a short peptide sequence that is easily recognized by tag-specific antibodies. Due to their small size, epitope tags do not affect the tagged protein's biochemical properties. Most often sequences encoding the epitope tag are included with target DNA at the time of cloning to produce fusion proteins containing the epitope tag sequence. This allows anti-epitope tag antibodies to serve as universal detection reagents for any tag containing protein produced by recombinant means. This means that anti-epitope tag antibodies are a useful alternative to generating specific antibodies to identify, immunoprecipitate or immunoaffinity purify a recombinant protein. The anti-epitope tag antibody is usually functional in a variety of antibody-dependent experimental procedures. Expression vectors producing epitope tag fusion proteins are available for a variety of host expression systems including bacteria, yeast, insect and mammalian cells.
Synonyms:	mouse anti-6X His Tag Antibody, anti-HIS, HIS Antibody, 6X His Tag Antibody, HHHHHH epitope tag antibody
Host Species:	Mouse
Clonality:	Monoclonal
Clone ID:	33D10.D2.G8
Format:	IgG1

Target Details

Reactivity:	His-Tag
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Immunogen Type:	Conjugated Peptide
Immunogen:	This His antibody was produced in mice by repeated immunizations with 6X His epitope tag peptide H-H-H-H-H-H conjugated to KLH using maleimide.
Purity/Specificity:	6X His antibody was purified from concentrated tissue culture supernate by Protein A chromatography and is directed against the 6X His motif and is useful in determining its presence in various assays. This polyclonal anti-6X His tag antibody detects over-expressed proteins containing the 6X His epitope tag. To date, this antibody has reacted with all His tagged proteins so far tested. In western blotting of bacterial extracts, the antibody does not cross-react with endogenous proteins. The antibody recognizes the His-tag (His-His-His-His-His-His) fused to either the amino- or carboxy- termini of targeted proteins in transfected or transformed cells.

Application Details

Tested Applications:	ELISA, WB
Application Note:	Anti-6X His is optimally suited for monitoring expression of His-tagged fusion proteins. As such, Anti-6X His/6XHIS can be used to identify fusion proteins that contain the 6X His epitope. The antibody recognizes the His tag fused either to the amino- or carboxy- termini of targeted proteins. This antibody has been tested by ELISA and western blotting against both the immunizing peptide and His-containing recombinant proteins. Although not tested, this antibody is likely functional for immunoprecipitation and immunocytochemistry.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:20,000 - 1:100,000
FC:	User Optimized
IHC:	User Optimized
IP:	1:200
WB:	1:2,000 - 1:10,000

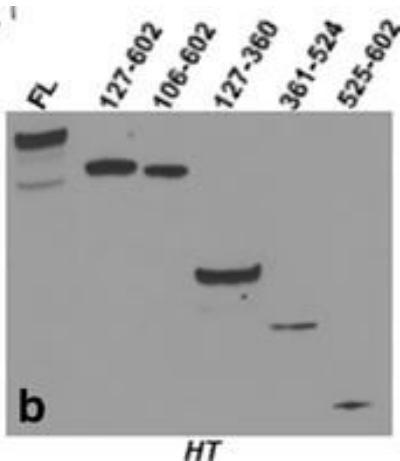
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	5.684 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	None

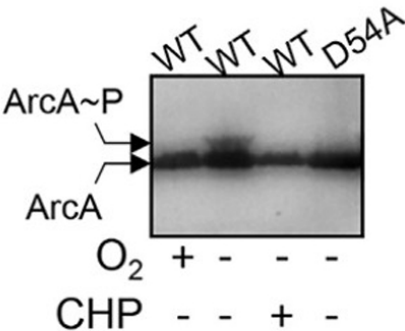
Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store 6X His Tag Antibody at -20° C or below prior to opening. This vial contains a relatively low volume of reagent (25 µL). To minimize loss of volume dilute 1:10 by adding 225 µL of the buffer stated above directly to the vial. Recap, mix thoroughly and briefly centrifuge to collect the volume at the bottom of the vial. Use this intermediate dilution when calculating final dilutions as recommended below. Store the vial at -20°C or below after dilution. Avoid cycles of freezing and thawing.
Expiration:	Expiration date is three (3) months from date of receipt.

Images



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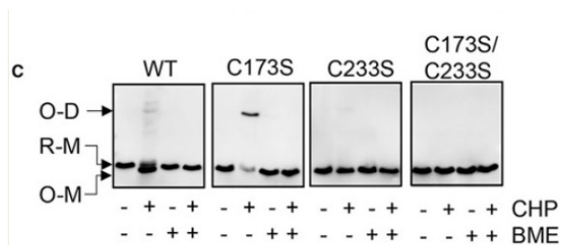


Western Blot

Characterization of purity and antigenicity of CKAP4 constructs. a SDS-PAGE of normalized amounts of purified CKAP4 constructs followed by silver staining. b–f Western blots using anti-His tag, anti-CKAP4 (G1/296), anti-CKAP4 (epitope region: 216–265), anti-CKAP4 (epitope region: 375–425), and anti-CKAP4 (epitope region: 552–602) primary antibody, respectively. Separate blots were used for each antibody. Molecular weight standards are indicated on left and right Figure provided by CiteAb. Source: BMC Biochem, PMID: 28893174.

Western Blot

ROS effects on ArcA phosphorylation and activity. (A) ArcA phosphorylation. *V. cholerae* overexpressing N-terminally His-tagged ArcAWT or ArcAD54A were grown aerobically (O₂+), microaerobically (O₂-), or microaerobically in the presence of 50 µM CHP. Whole-cell lysates were analyzed by Phos-Tag gel electrophoresis followed by anti-His6 western blotting to visualize ArcA phosphorylation. Figure 4. PMID: 34936880



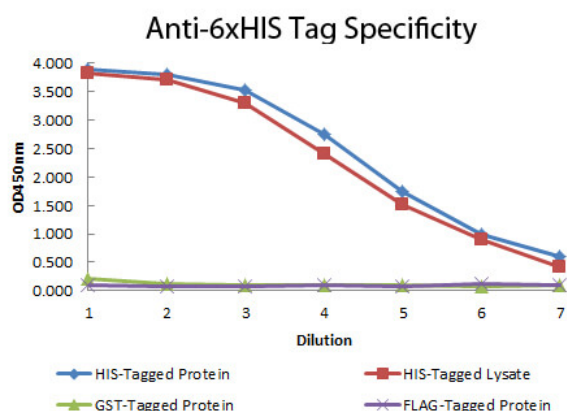
Western Blot

ArcA thiol oxidation induces intramolecular disulfide bond formation and enhances ArcA-ArcA interaction.

(C) *V. cholerae* containing overexpressed His6-tagged ArcA variants were grown under the virulence-inducing condition. When indicated, 50 μ M CHP was added during ArcA induction. Cell lysates (normalized by optical density at 600 nm [OD₆₀₀]) with or without 100 mM BME were separated on a non-reducing 12% SDS-polyacrylamide gel followed by western blotting analysis using anti-His6 antibody; O-M, oxidized monomer; R-M, reduced monomer; O-D, oxidized dimer.

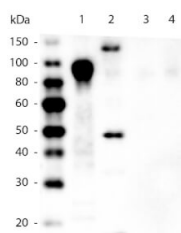
Figure 5.

PMID: 34936880



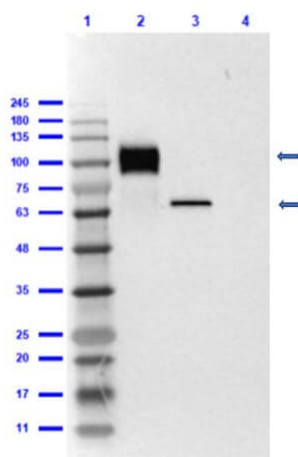
ELISA

ELISA of Mouse anti-6xHIS Tag Antibody. Antigen: HIS-tagged purified protein and *E. coli* cell lysates expressing HIS-Tagged construct, GST- and RON-tagged purified proteins. Coating amount: 0.15 μ g per well. Primary antibody: 6xHIS Tag antibody at 100 μ g/mL. Dilution series: 2-fold. Mid-point concentration: 200 ng/mL. Secondary antibody: Peroxidase mouse secondary antibody at 1:10,000. Substrate: TMB (p/n TMBE-1000).



Western Blot

Western Blot of Mouse anti-6xHIS Tag Antibody. Lane 1: 100 ng Purified histidine-tagged recombinant protein. Lane 2: 200 ng *E. coli* cell lysate containing histidine-tagged expression construct. Lane 3: 100 ng Purified GST-tagged recombinant protein. Lane 4: 100 ng Purified FLAG-tagged recombinant protein. Primary antibody: Mouse anti-6xHIS Tag antibody at 1:5,000 overnight at 4°C. Secondary antibody: Peroxidase mouse secondary antibody at 1:20,000 for 30 min at RT. Block: 5% BLOTTO for 1 hr at RT.



Western Blot

Western Blot of Mouse Anti-6X HIS Epitope Tag Antibody.
 Lane 1: Opal Prestained Molecular Weight Marker (p/n MB-210-0500). Lane 2: rec EGF -C-term HIS tag [0.02µg] [+]. Lane 3: rec Luciferase N-term HIS tag [0.10µg] [+]. Lane 4: Zic1 - C-myc/DDK Tag [0.10µg] [-]. Primary Antibody: Mouse Anti-6X HIS at 1.0µg/mL overnight at 2-8°C. Secondary Antibody: Rabbit Anti-Mouse HRP (p/n 610-403-C46) at 1:40,000 for 30mins at RT. Block: BlockOut Buffer (p/m MB-073) for 1hr at RT. Bands are recognized at the expected MW. Exposure: 2sec.

References

- Zhou Y et al. Thiol-based functional mimicry of phosphorylation of the two-component system response regulator ArcA promotes pathogenesis in enteric pathogens. *Cell Rep.* (2021)
- Kao CH et al. Growth-regulated Hsp70 phosphorylation regulates stress responses and prion maintenance. *Mol Cell Biol.* (2020)
- Romero-Rodriguez A et al. The Clostridioides difficile Cysteine-Rich Exosporium Morphogenetic Protein, CdeC, Exhibits Self-Assembly Properties That Lead to Organized Inclusion Bodies in Escherichia coli. *mSphere.* (2020)
- Chavda et al. Antiproliferative factor (APF) binds specifically to sites within the cytoskeleton-associated protein 4 (CKAP4) extracellular domain. *BMC Biochemistry* (2017)
- Ramón-García S, Ng C, Jensen PR, et al. WhiB7, an Fe-S-dependent transcription factor that activates species-specific repertoires of drug resistance determinants in actinobacteria. *J Biol Chem.* (2013)

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