abcam

Product datasheet

Anti-6X His tag® antibody [HIS.H8] ab18184

★★★★★ 32 Abreviews 312 References 5 Images

Overview

Product name Anti-6X His tag® antibody [HIS.H8]

Description Mouse monoclonal [HIS.H8] to 6X His tag®

Host species Mouse

Specificity Recognizes His-tagged recombinant proteins or His-tagged proteins overexpressed in cells.

ab18184 reacts to recombinant proteins containing the 6X His tag® or 10X His tag® fused to

either the amino or carboxy terminus.

Tested applications Suitable for: ICC/IF, ELISA, Dot blot, WB, IP

Species reactivity Reacts with: Species independent

Immunogen Synthetic peptide corresponding to 6X His tag®.

General notesThis product was changed from ascites to tissue culture supernatant on 5th February 2018.

Please note that the dilutions may need to be adjusted accordingly. If you have any questions,

please do not hesitate to contact our scientific support team.

HIS-TAG® is a trademark of EMD Biosciences, Inc.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

Storage buffer pH: 7.20

Preservative: 0.05% Sodium azide

Constituent: PBS

Purity Protein A purified

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Purification notes Purified from TCS by Protein A affinity chromatography

ClonalityMonoclonalClone numberHIS.H8IsotypeIgG2b

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab18184 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF	★★★★ <u>(3)</u>	1/200.
ELISA	*** <u>*</u>	Use at an assay dependent concentration.
Dot blot		Use at an assay dependent concentration.
WB	★★★★★ (21)	1/1000.
IP	★★★★ ☆ <u>(5)</u>	1/200.

Target

Relevance The H-H-H-H motif is used as a tag on many recombinant proteins to facilitate purification.

His-tags can be fused to the amino- or carboxy- termini of proteins in transfected or transformed

cells.

Cellular localization Depends upon the localization of the parent protein tagged with hexahistidine.

Images



Immunoprecipitation - Anti-6X His tag® antibody [HIS.H8] (ab18184)

This image is courtesy of an Abreview submitted by Mr Chris Tracy

Immunoprecipitation of His/FLAG tagged PDCD5 using ab18184 at 1/100 dilution.

Lane 1 : MW Ladder (from top 120, 85, 50, 34, 25, 19 kDa) **Lanes 2-5 :** Immunoprecipitation of His/FLAG tagged PDCD5 using ab18184 from mixture of ~50 ng purified His/FLAG tagged PDCD5 and 15 μ L RRL in 300 μ L buffer.

Lanes 6-9: Purified PDCD5 from E. coli with ~60-70 ng protein.

The lowest band in each lane is \sim 20 kDa and is PDCD5. The upper bands in lanes 2-5 are the heavy chain (\sim 50 kDa, runs as doublet) and light chain (\sim 25 or 26 kDa).

Western Blot was performed with a mouse anti-FLAG primary antibody at 1/2000 dilution. An IRDye®800CW-conjugated goat anti-mouse IgG was used as a secondary antibody.

All lanes : Anti-6X His tag® antibody [HIS.H8] (ab18184) at 1/5000 dilution

Lane 1 : ARPE-19 whole cell lysate - non-transfected

Lane 2: ARPE-19 whole cell lysate - transfected with rRab27a-His

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : HRP-conjugated goat anti-mouse lgG polyclonal at 1/5000 dilution

Developed using the ECL technique.

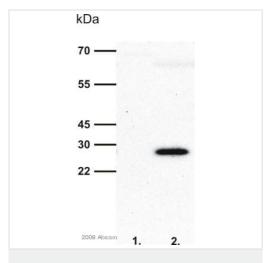
Performed under reducing conditions.

Exposure time: 5 minutes

Blocked with 5% milk for 30 minutes at 25°C.

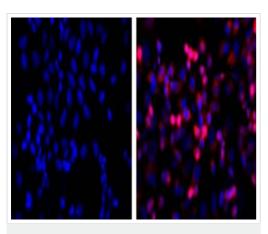
Incubated with the primary antibody for 16 hours at $4\,^{\circ}\text{C}$ in 1X PBS/Tween, 5% milk.

LEFT: untransfected control; RIGHT: anti-His (in red) on His-tagged fusion proteins in HEK293 cells. Both counterstained with DAPI (in blue).

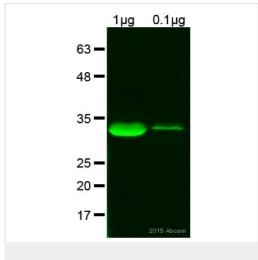


Western blot - Anti-6X His tag® antibody [HIS.H8] (ab18184)

This image is courtesy of an Abreview submitted by Vladimir Milenkovic



Immunocytochemistry/ Immunofluorescence - Anti-6X His tag® antibody [HIS.H8] (ab18184)



Western blot - Anti-6X His tag® antibody [HIS.H8] (ab18184)

This image is courtesy of an Abreview submitted by Rachel Loh

Immunocytochemistry/ Immunofluorescence - Anti-

6X His tag® antibody [HIS.H8] (ab18184)
This image is courtesy of an anonymous abreview.

All lanes : Anti-6X His tag® antibody [HIS.H8] (ab18184) at 1/1000 dilution

Lane 1: 6x His-tagged protein expressed in BL21 (pET15b) at 1

Lane 2: 6x His-tagged protein expressed in BL21 (pET15b) at 0.1 µg

Secondary

All lanes : IRDye® 800CW goat anti-mouse IgG (H+L) polyclonal at 1/5000 dilution

Performed under reducing conditions.

Exposure time: 5 minutes

ab18184 at a 1/1000 dilution staining 6X His tag in transfected human HEK293 cells by Immunocytochemistry/
Immunofluorescence incubated for 12 hours at 22°C. PFA fixed.
Permeabilized using 0.1% Triton X-100 in PBS. Blocked using 3% BSA for 1 hour at 22°C. Secondary used at a 1/1000 dilution polyclonal Goat anti-mouse IgG (H+L) conjugated to Alexa Fluor 488 (green). DAPI staining (blue).

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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