abcam

Product datasheet

Anti-6X His tag® antibody [4D11] ab5000

Overview

Product name Anti-6X His tag® antibody [4D11]

Description Mouse monoclonal [4D11] to 6X His tag®

Host species Mouse

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

Species reactivity Reacts with: Species independent

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Epitope This antibody was shown to recognize an epitope (HHHHHHGS) encoded by the polylinker of

several expression vectors (including pDS:RBSII, pQE-30, and pQE-50).

General notes

This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact orders@abcam.com.

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). Add glycerol to a final volume of 50% for

extra stability and aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide Constituents: 6.97% L-Arginine, PBS

Purity Protein G purified

Clonality Monoclonal

Clone number 4D11

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MyelomaNS1IsotypeIgG2aLight chain typekappa

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab5000 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	
AP		Use at an assay dependent concentration.	
Dot blot		Use at an assay dependent concentration.	
ELISA		Use at an assay dependent concentration.	
WB	★★★★ <u>(2)</u>	1/1000.	
IP	★★★★ ☆ (1)	Use at an assay dependent concentration. See Abreview.	
ICC/IF	****(1)	Use at an assay dependent concentration.	
Flow Cyt		Use at an assay dependent concentration. <u>ab170191</u> - Mouse monoclonal lgG2a, is suitable for use as an isotype control with this antibody.	

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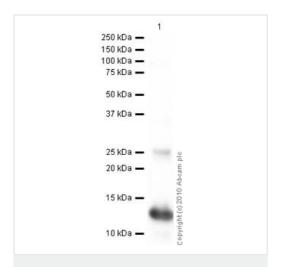
Relevance The H-H-H-H motif is used as a tag on many recombinant proteins to facilitate purification.

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



Western blot - Anti-6X His tag® antibody [4D11] (ab5000)

Anti-6X His tag® antibody [4D11] (ab5000) at 1/1000 dilution + 6X His Tagged Protein at 1 μg

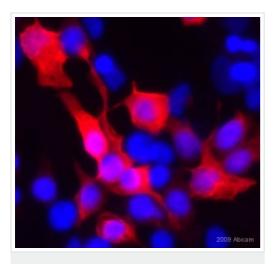
Secondary

Goat polyclonal to Mouse IgG - H&L - Pre-Adsorbed (HRP) (ab65485) at 1/3000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

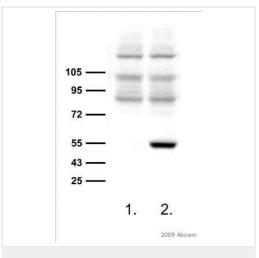
Exposure time: 30 seconds



Immunocytochemistry/ Immunofluorescence - Anti-6X His tag® antibody [4D11] (ab5000)

This image is courtesy of an Abreview submitted by Dr Vladimir Mlenkovic

ab5000 staining Human HEK 293 cells transfected with CACNB3-His by ICC/IF. Cells were PFA fixed and permeabilized in 0.5% Triton X-100 prior to blocking in 5% serum for 20 minutes at 25°C. The primary antibody (1/250 in 1% goat serum, 0.1% TX100, 1X PBS) was incubated with the sample for 16 hours at 4°C. An Alexa Fluor[®] 546 conjugated goat anti-mouse antibody (1/500) was used as the secondary. DAPI was used for nuclei staining (blue).



Western blot - Anti-6X His tag® antibody [4D11] (ab5000)

This image is courtesy of an Abreview submitted by Dr Vladimir Mlenkovic

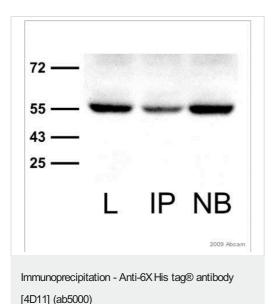
All lanes : Anti-6X His tag® antibody [4D11] (ab5000) at 1/2000 dilution

Lane 1: Lysates prepared from HEK-293 cells

Lane 2: Lysates prepared from HEK-293 cells transfected with Beta3-His (CACNB3)

Secondary

All lanes : HRP-conjugated goat anit-mouse IgG polyclonal at 1/5000 dilution



This image is courtesy of an Abreview submitted by Dr

Madimir Milenkovic

ab5000 Immunoprecipitate in human HEK293 whole cell lysate. 25µg of cell lysate incubated with primary antibody (undiluted) and matrix (Protein G) for 16 hours at 4°C. For western blotting an undiluted HRP-Goat anti-rabbit lgG polyclonal was used. Beta3-His fusion protein was immunoprecipitated using anti His Ab, and specific beta3a band (56kDa) was detected using anti beta3 Ab

Lane 1. Lysate of HEK293 cells expressing beta3-His (CACNB3)

Lane 2. IP with anti His Ab

Lane 3. Non bound fraction

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