

Product datasheet

Anti-Bub1 antibody [14H5] ab181438

KO VALIDATED

1 Image

Overview

Product name	Anti-Bub1 antibody [14H5]
Description	Mouse monoclonal [14H5] to Bub1
Host species	Mouse
Tested applications	Suitable for: WB, IP, FM
Species reactivity	Reacts with: Human
Immunogen	Recombinant fragment corresponding to Human Bub1 aa 281-419 (internal sequence). Genbank: NP_004327.1 Sequence: ANAFEEQLLK QKMDELHKKL HQVVETSHED LPASQERSEV NPARMGPSVG SQQELRAPCL PVTYQQTPVN MEKNPREAPP VVPPLANAIS AALVSPATSQ SIAPPVPLKA QTVTDSMFAV ASKDAGCVNK STHEFKPQS Database link: Q53QE4 Run BLAST with Run BLAST with
Positive control	HeLa whole cell lysates prepared using a RIPA buffer.

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 long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 0.88% Sodium chloride, 0.27% Potassium phosphate Sterile filtered
Purity	Protein A purified
Purification notes	ab181438 was purified from tissue culture supernatant by chromatography.

Clonality	Monoclonal
Clone number	14H5
Isotype	IgG1

Applications

Our [Abpromise guarantee](#) covers the use of **ab181438** 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
WB		1/500 - 1/2000. Predicted molecular weight: 160 kDa.
IP		1/200.
FM		1/500 - 1/2000. It is recommended to use cells grown on cover slips fixed with 3.5% paraformaldehyde in PBS at pH 6.8 OR 100% methanol at -20° C. Permeabilize fixed cells with 0.5% Triton X-100.

Target

Function

Serine/threonine-protein kinase that performs 2 crucial functions during mitosis: it is essential for spindle-assembly checkpoint signaling and for correct chromosome alignment. Has a key role in the assembly of checkpoint proteins at the kinetochore, being required for the subsequent localization of CENPF, BUB1B, CENPE and MAD2L1. Required for the kinetochore localization of PLK1. Plays an important role in defining SGOL1 localization and thereby affects sister chromatid cohesion. Acts as a substrate for anaphase-promoting complex or cyclosome (APC/C) in complex with its activator CDH1 (APC/C-Cdh1). Necessary for ensuring proper chromosome segregation and binding to BUB3 is essential for this function. Can regulate chromosome segregation in a kinetochore-independent manner. Can phosphorylate BUB3. The BUB1-BUB3 complex plays a role in the inhibition of APC/C when spindle-assembly checkpoint is activated and inhibits the ubiquitin ligase activity of APC/C by phosphorylating its activator CDC20. This complex can also phosphorylate MAD1L1. Kinase activity is essential for inhibition of APC/CCDC20 and for chromosome alignment but does not play a major role in the spindle-assembly checkpoint activity. Mediates cell death in response to chromosome missegregation and acts to suppress spontaneous tumorigenesis.

Tissue specificity

High expression in testis and thymus, less in colon, spleen, lung and small intestine. Expressed in fetal thymus, bone marrow, heart, liver, spleen and thymus. Expression is associated with cells/tissues with a high mitotic index.

Sequence similarities

Belongs to the protein kinase superfamily. Ser/Thr protein kinase family. BUB1 subfamily. Contains 1 BUB1 N-terminal domain. Contains 1 protein kinase domain.

Domain

The KEN box is required for its ubiquitination and degradation. BUB1 N-terminal domain directs kinetochore localization and binding to BUB3.

Post-translational modifications

Phosphorylated upon DNA damage, probably by ATM or ATR. Upon spindle-assembly checkpoint activation it is hyperphosphorylated and its kinase activity toward CDC20 is stimulated. Phosphorylation at Thr-609 is required for interaction with PLK1, phosphorylation at

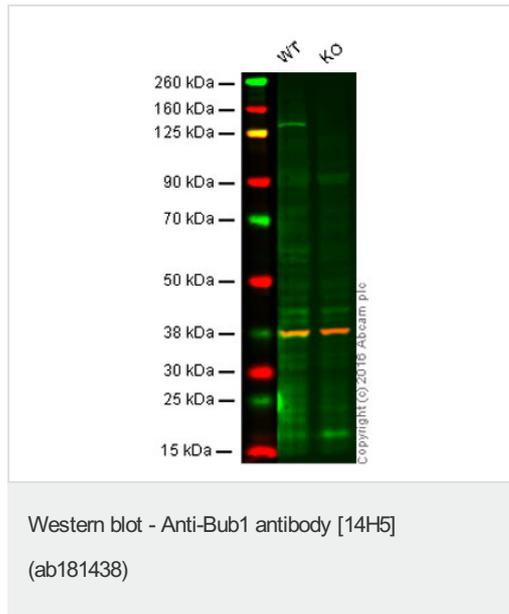
this site probably creates a binding site for the POLO-box domain of PLK1, thus enhancing the PLK1-BUB1 interaction.

Ubiquitinated and degraded during mitotic exit by APC/C-Cdh1.

Cellular localization

Nucleus. Chromosome > centromere > kinetochore. Nuclear in interphase cells. Accumulates gradually during G1 and S phase of the cell cycle, peaks at G2/M, and drops dramatically after mitosis. Localizes to the outer kinetochore. Kinetochore localization is required for normal mitotic timing and checkpoint response to spindle damage and occurs very early in prophase. AURKB, CASC5 and INCENP are required for kinetochore localization.

Images



Lane 1: Wild-type HAP1 cell lysate (40 µg)

Lane 2: Bub1 knockout HAP1 cell lysate (40 µg)

Lanes 1 - 2: Merged signal (red and green). Green - ab181438 observed at 125 kDa. Red - loading control, ab181602, observed at 37 kDa.

ab181438 was shown to recognize Bub1 when Bub1 knockout samples were used, along with additional cross-reactive bands. Wild-type and Bub1 knockout samples were subjected to SDS-PAGE. Ab181438 and ab181602 (loading control to GAPDH) were diluted at 1/500 and 1:10,000 dilution respectively and incubated overnight at 4C. Blots were developed with IRDye® 800CW Goat anti-Mouse IgG (H + L) and IRDye® 680 Goat anti-Rabbit IgG (H + L) secondary antibodies at 1:10,000 dilution for 1 hour at room temperature before imaging.

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

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <https://www.abcam.com/abpromise> or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors