abcam

Product datasheet

Anti-RBX1 antibody [EPR6850(B)] - BSA and Azide free ab248556



8 Images

Overview

Product name Anti-RBX1 antibody [EPR6850(B)] - BSA and Azide free

Description Rabbit monoclonal [EPR6850(B)] to RBX1 - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), IHC-P, ICC/IF, IP, WB

Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control IP: Jurkat whole cell lysate. Flow Cyt: Jurkat cells. ICC/IF: HeLa cells. IHC-P: Rat and mouse

breast tissue. Human lung carcinoma tissue. WB: Jurkat whole cell lysate. Mouse and rat heart

lysate.

General notes ab248556 is the carrier-free version of ab133565.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

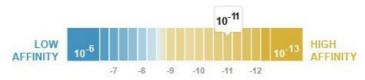
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Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Do Not Freeze.

Dissociation constant (K_D) $K_D = 2.18 \times 10^{-11} M$



Learn more about K_D

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR6850(B)

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab248556 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
Flow Cyt (Intra)		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 13 kDa (predicted molecular weight: 12 kDa).

Target

Function

E3 ubiquitin ligase component of multiple cullin-RING-based E3 ubiquitin-protein ligase complexes which mediate the ubiquitination and subsequent proteasomal degradation of target proteins, including proteins involved in cell cycle progression, signal transduction, transcription and transcription-coupled nucleotide excision repair. The functional specificity of the E3 ubiquitin-

protein ligase complexes depends on the variable substrate recognition components. As a component of the CSA complex promotes the ubiquitination of ERCC6 resulting in proteasomal degradation. Through the RING-type zinc finger, seems to recruit the E2 ubiquitination enzyme, like CDC34, to the complex and brings it into close proximity to the substrate. Probably also stimulates CDC34 autoubiquitination. May be required for histone H3 and histone H4 ubiquitination in response to ultraviolet and for subsequent DNA repair. Promotes the neddylation

Tissue specificity Widely expressed.

Pathway Protein modification; protein ubiquitination.

Sequence similarities Belongs to the RING-box family.

Contains 1 RING-type zinc finger.

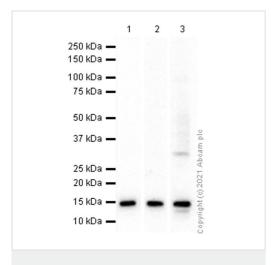
DomainThe RING-type zinc finger domain is essential for ubiquitin ligase activity. It coordinates an

of CUL1, CUL2, CUL4 and CUL4 via its interaction with UBE2M.

additional third zinc ion.

Cellular localization Cytoplasm. Nucleus.

Images



Western blot - Anti-RBX1 antibody [EPR6850(B)] - BSA and Azide free (ab248556)

All lanes : Anti-RBX1 antibody [EPR6850(B)] (**ab133565**) at 1/5000 dilution

Lane 1 : Jurkat (Human T cell leukemia T lymphocyte) whole cell

lysate

Lane 2 : Mouse heart lysate

Lane 3 : Rat heart lysate

Lysates/proteins at 15 µg per lane.

Secondary

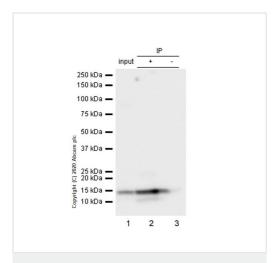
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 12 kDa

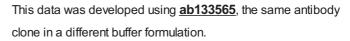
This data was developed using <u>ab133565</u>, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.

Image produced using the purified version.



Immunoprecipitation - Anti-RBX1 antibody
[EPR6850(B)] - BSA and Azide free (ab248556)



Purified <u>ab133565</u> at 1:20 dilution ($1\mu g$) immunoprecipitating RBX1 in Jurkat whole cell lysate.

Lane 1 (input): Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate $10\mu g$.

Lane 2 (+): ab133565 + Jurkat whole cell lysate.

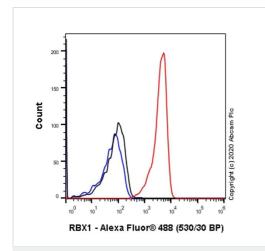
Lane 3 (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of <u>ab133565</u> in Jurkat whole cell lysate.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) (1:1000 dilution) was used for Western blotting.

Blocking Buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM/TBST.

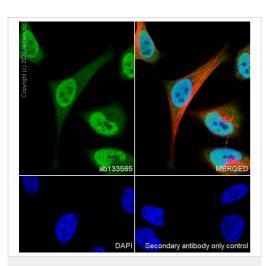
Observed band size: 13 kDa



Flow Cytometry (Intracellular) - Anti-RBX1 antibody [EPR6850(B)] - BSA and Azide free (ab248556)

This data was developed using <u>ab133565</u>, the same antibody clone in a different buffer formulation.

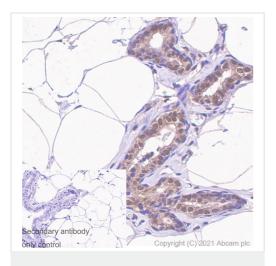
Flow Cytometry analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labelling RBX1 with Purified ab133565 at 1:20 dilution (10 µg/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor® 488, ab150077) secondary antibody was used at 1:2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunocytochemistry/ Immunofluorescence - Anti-RBX1 antibody [EPR6850(B)] - BSA and Azide free (ab248556)

This data was developed using <u>ab133565</u>, the same antibody clone in a different buffer formulation.

Immunocytochemistry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling RBX1 with Purified **ab133565** at 1:50 dilution (3.9 μg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 μg/ml). Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1:1000 (2 μg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

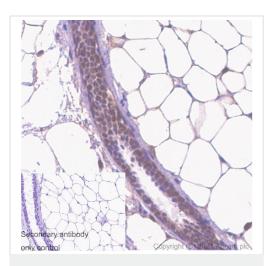


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody

[EPR6850(B)] - BSA and Azide free (ab248556)

This data was developed using <u>ab133565</u>, the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat breast tissue sections labeling RBX1 with Purified <u>ab133565</u> at 1:2000 dilution (0.099 µg/mL). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.

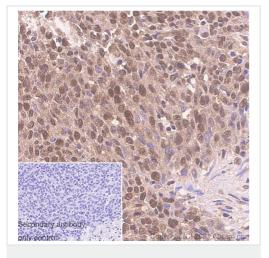


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody

[EPR6850(B)] - BSA and Azide free (ab248556)

This data was developed using <u>ab133565</u>, the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse breast tissue sections labeling RBX1 with Purified <u>ab133565</u> at 1:2000 dilution (0.099 µg/mL). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.

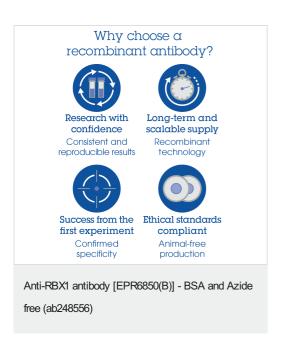


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody

[EPR6850(B)] - BSA and Azide free (ab248556)

This data was developed using <u>ab133565</u>, the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human lung carcinoma tissue sections labeling RBX1 with Purified <u>ab133565</u> at 1:2000 dilution (0.099 µg/mL). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.



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