abcam

Product datasheet

Anti-Aurora B antibody [EP1009Y] ab45145

Recombinant RabMAb

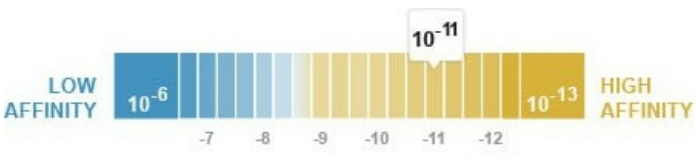
1 Abreviews 24 References 7 Images

Overview

Product name	Anti-Aurora B antibody [EP1009Y]
Description	Rabbit monoclonal [EP1009Y] to Aurora B
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), ICC/IF, WB, IHC-P, IP
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide within Human Aurora B aa 1-100 (N terminal). The exact sequence is proprietary.
Positive control	IP: HeLa cell lysate WB: HeLa cell lysate IHC: human endometrium carcinoma ICC: HeLa cells
General notes	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**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Dissociation constant (K_D)	$K_D = 5.50 \times 10^{-11}$ M



Learn more about K_D

Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP1009Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab45145 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)		1/300.
ICC/IF		1/20. For unpurified version use 1/100 - 1/150 dilution
WB	★★★★☆ (1)	Use at an assay dependent concentration. Predicted molecular weight: 39 kDa. For unpurified version use at 1/50000 dilution
IHC-P		1/200. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. For unpurified version use at 1/250-500
IP		Use at an assay dependent concentration. For unpurified version use at 1/20 dilution

Target

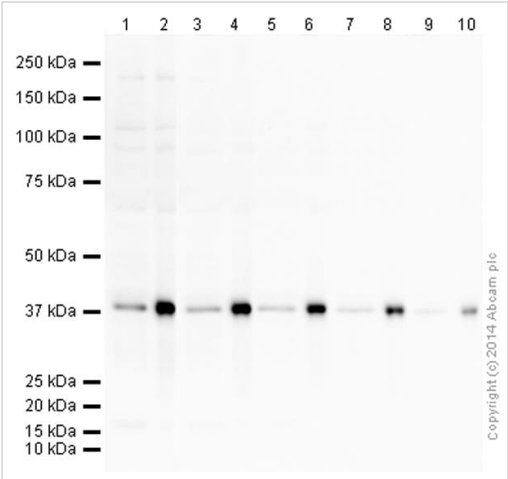
Function	May be directly involved in regulating the cleavage of polar spindle microtubules and is a key regulator for the onset of cytokinesis during mitosis. Component of the chromosomal passenger complex (CPC), a complex that acts as a key regulator of mitosis. The CPC complex has essential functions at the centromere in ensuring correct chromosome alignment and segregation and is required for chromatin-induced microtubule stabilization and spindle assembly. Phosphorylates 'Ser-10' and 'Ser-28' of histone H3 during mitosis. Required for kinetochore localization of BUB1 and SGOL1. Interacts with INCENP.
Tissue specificity	High level expression seen in the thymus. It is also expressed in the spleen, lung, testis, colon, placenta and fetal liver. Expressed during S and G2/M phase and expression is up-regulated in cancer cells during M phase.
Involvement in disease	Note=Disruptive regulation of expression is a possibile mechanism of the perturbation of chromosomal integrity in cancer cells through its dominant-negative effect on cytokinesis.
Sequence similarities	Belongs to the protein kinase superfamily. Ser/Thr protein kinase family. Aurora subfamily. Contains 1 protein kinase domain.
Post-translational modifications	Ubiquitinated by different BCR (BTB-CUL3-RBX1) E3 ubiquitin ligase complexes. Ubiquitinated by the BCR(KLHL9-KLHL13) E3 ubiquitin ligase complex, ubiquitination leads to removal from

mitotic chromosomes and is required for cytokinesis. During anaphase, the BCR(KLHL21) E3 ubiquitin ligase complex recruits the CPC complex from chromosomes to the spindle midzone and mediates the ubiquitination of AURKB. Ubiquitination of AURKB by BCR(KLHL21) E3 ubiquitin ligase complex may not lead to its degradation by the proteasome.

Cellular localization

Nucleus. Chromosome. Chromosome > centromere. Cytoplasm > cytoskeleton > spindle. Localizes on chromosome arms and inner centromeres from prophase through metaphase and then transferring to the spindle midzone and midbody from anaphase through cytokinesis. Colocalized with gamma tubulin in the mid-body.

Images



Western blot - Anti-Aurora B antibody [EP1009Y] (ab45145)

Lanes 1-2 : Anti-Aurora B antibody [EP1009Y] (ab45145) at 1/1000 dilution
Lanes 3-4 : Anti-Aurora B antibody [EP1009Y] (ab45145) at 1/5000 dilution
Lanes 5-6 : Anti-Aurora B antibody [EP1009Y] (ab45145) at 1/10000 dilution
Lanes 7-8 : Anti-Aurora B antibody [EP1009Y] (ab45145) at 1/50000 dilution
Lanes 9-10 : Anti-Aurora B antibody [EP1009Y] (ab45145) at 1/75000 dilution

Lanes 1 & 3 & 5 & 7 & 9 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate
Lanes 2 & 4 & 6 & 8 & 10 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate Nocodozole Stimulated

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Anti-Rabbit IgG VHH Single Domain (HRP) (**ab191866**) at 1/10000 dilution

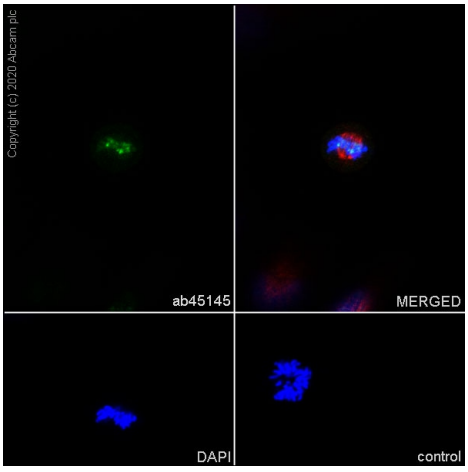
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 39 kDa
Observed band size: 39 kDa

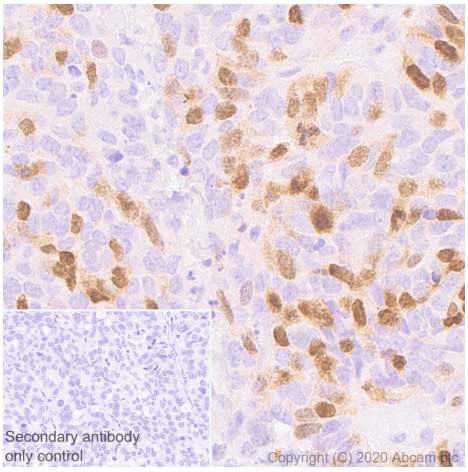
Exposure time: 8 minutes

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with ab45145 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution **ab133406**



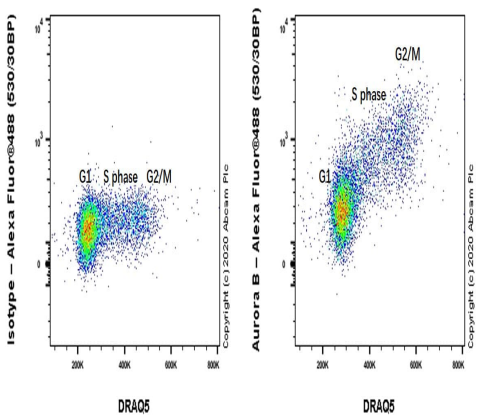
Immunocytochemistry/ Immunofluorescence - Anti-Aurora B antibody [EP1009Y] (ab45145)

Immunocytochemistry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Aurora B with Purified ab45145 at 1:50 dilution (5.4 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 IgG (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.



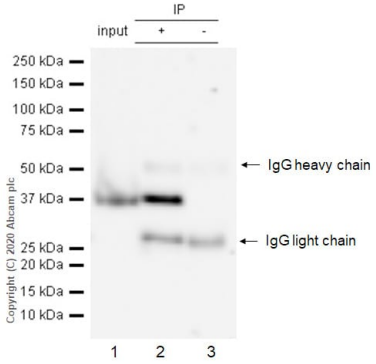
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Aurora B antibody [EP1009Y] (ab45145)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human endometrium carcinoma tissue sections labeling Aurora B with purified ab45145 at 1/200 dilution (1.35 µg/mL). Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0). The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



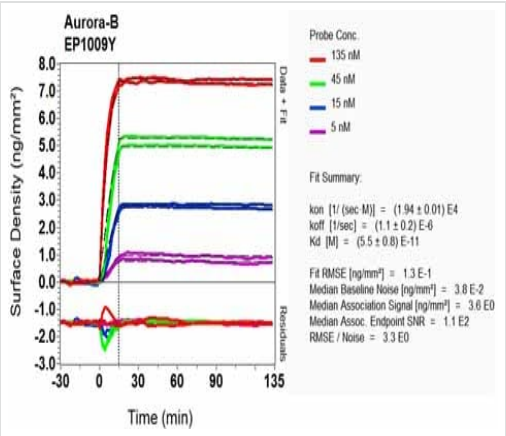
Flow Cytometry (Intracellular) - Anti-Aurora B antibody [EP1009Y] (ab45145)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Aurora B with Purified ab45145 at 1/300 dilution (0.1 µg/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488 ,**ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Left). Unlabeled control - /.



Immunoprecipitation - Anti-Aurora B antibody [EP1009Y] (ab45145)

Purified ab45145 at 1/20 dilution (1µg) immunoprecipitating Aurora B in HeLa whole cell lysate.
Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10µg
Lane 2 (+): ab45145 + HeLa whole cell lysate.
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of ab45145 in HeLa whole cell lysate.
VeriBlot for IP Detection Reagent (HRP) (**ab131366**) (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: 39 kDa

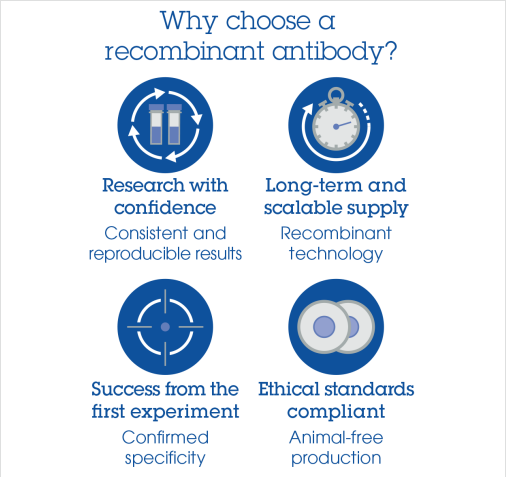


OI-RD Scanning - Anti-Aurora B antibody [EP1009Y] (ab45145)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D



Anti-Aurora B antibody [EP1009Y] (ab45145)

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