abcam

Product datasheet

Anti-PLK1 antibody [35-206] ab17056

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Overview

Product name	Anti-PLK1 antibody [35-206]
Description	Mouse monoclonal [35-206] to PLK1
Host species	Mouse
Tested applications	**Suitable for:** Flow Cyt, ICC/IF, IHC-P, IP, WB, ICC
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	His-PLK1 full length purified from Sf9 cells.
Epitope	aa330-370.
General notes	This antibody clone is manufactured by Abcam. If you require this antibody in a particular buffer formulation or a particular conjugate for your experiments, please contact orders@abcam.com or you can find further information here.

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: 7.40 Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine
Purity	IgG fraction
Clonality	Monoclonal
Clone number	35-206
Isotype	IgG2b

Applications

Our Abpromise guarantee covers the use of **ab17056** 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		Use 1µg for 10^6 cells. ab170192 - Mouse monoclonal IgG2b, is suitable for use as an isotype control with this antibody.
ICC/IF	★★★★☆	Use at an assay dependent concentration.
IHC-P	★★★★☆	Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
WB	★★★★★	Use a concentration of 1 µg/ml. Detects a band of approximately 66 kDa (predicted molecular weight: 67 kDa). This clone is superior to clone 36-298 (ab17057) in Western blotting and IP, but suffers from high background in IF and ICC.
ICC		Use at an assay dependent concentration.

Target

Function	Serine/threonine-protein kinase that performs several important functions throughout M phase of the cell cycle, including the regulation of centrosome maturation and spindle assembly, the removal of cohesins from chromosome arms, the inactivation of APC/C inhibitors, and the regulation of mitotic exit and cytokinesis. Required for recovery after DNA damage checkpoint and entry into mitosis. Required for kinetochore localization of BUB1B. Phosphorylates SGOL1. Required for spindle pole localization of isoform 3 of SGOL1 and plays a role in regulating its centriole cohesion function. Phosphorylates BORA, and thereby promotes the degradation of BORA. Contributes to the regulation of AURKA function. Regulates TP53 stability through phosphorylation of TOPORS.
Tissue specificity	Placenta and colon.
Sequence similarities	Belongs to the protein kinase superfamily. Ser/Thr protein kinase family. CDC5/Polo subfamily. Contains 2 POLO box domains. Contains 1 protein kinase domain.
Developmental stage	Accumulates to a maximum during the G2 and M phases, declines to a nearly undetectable level following mitosis and throughout G1 phase, and then begins to accumulate again during S phase.
Post-translational modifications	Catalytic activity is enhanced by phosphorylation of Thr-210. Phosphorylation at Thr-210 is first detected on centrosomes in the G2 phase of the cell cycle, peaks in prometaphase and gradually disappears from centrosomes during anaphase. Autophosphorylation and phosphorylation of Ser-137 may not be significant for the activation of PLK1 during mitosis, but may enhance catalytic activity during recovery after DNA damage checkpoint. Ubiquitinated by the anaphase promoting complex/cyclosome (APC/C) in anaphase and following DNA damage, leading to its degradation by the proteasome. Ubiquitination is mediated via its interaction with FZR1/CDH1. Ubiquitination and subsequent degradation prevents entry into mitosis and is essential to maintain an efficient G2 DNA damage checkpoint.
Cellular localization	Nucleus. Chromosome > centromere > kinetochore. Cytoplasm > cytoskeleton > centrosome. During early stages of mitosis, the phosphorylated form is detected on centrosomes and kinetochores. Localizes to the outer kinetochore. Presence of SGOL1 and interaction with the phosphorylated form of BUB1 is required for the kinetochore localization.

Images

Western blot - Anti-PLK1 antibody [35-206] (ab17056)

Anti-PLK1 antibody [35-206] (ab17056) at 1 µg/ml + A431 cell lysate at 20 µg

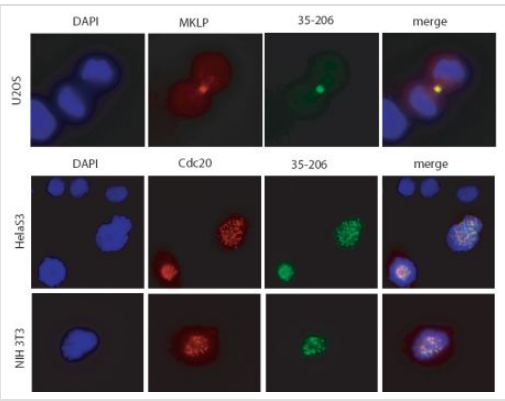
Secondary

Rabbit-anti mouse Alexa fluor(680) at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

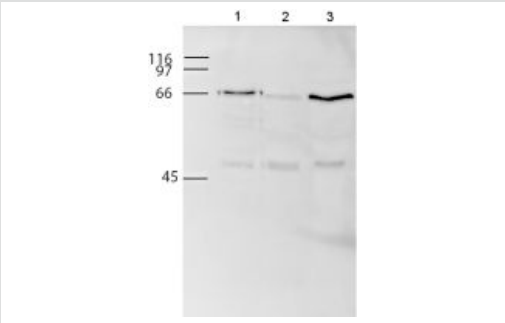
Predicted band size: 67 kDa
Observed band size: 66 kDa
why is the actual band size different from the predicted?



Immunocytochemistry/ Immunofluorescence - Anti-PLK1 antibody [35-206] (ab17056)

Immunofluoresence using ab17056 and either U2OS, HeLaS3 or NIH 3T3 cells.

Plk1 can be seen localising with MKLP in the U2OS cell and with Cdc20 in the HeLaS3 and NIH 3T3 cells shown.



Western blot - Anti-PLK1 antibody [35-206] (ab17056)

All lanes : Anti-PLK1 antibody [35-206] (ab17056)

Lane 1 : Recombinant PLK1
Lane 2 : U2OS lysate
Lane 3 : HeLaS3

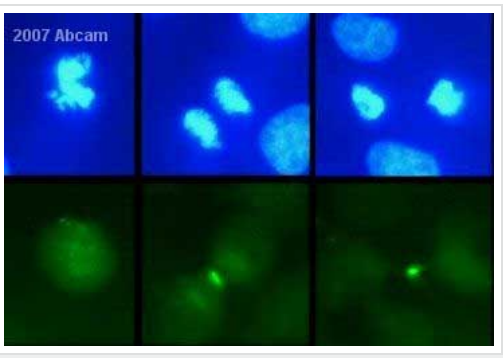
Performed under reducing conditions.

Predicted band size: 67 kDa
Observed band size: 66 kDa why is the actual band size different from the predicted?

Western blot using ab17056.

Lane 1: recombinant PLK1
Lane 2: U2OS lysate
Lane 3: HeLaS3 lysate

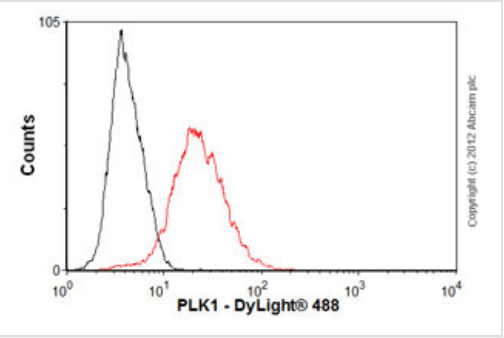
15% SDS-PAGE gel



Immunocytochemistry/ Immunofluorescence - Anti-PLK1 antibody [35-206] (ab17056)

This image is courtesy of an Abreview submitted by Melanie Adler

ab17056 at 1/100 staining human kidney tubular epithelial cells by ICC/IF. The cells were formaldehyde fixed, permeabilized with Triton X-100 and blocked with goat serum before incuabtion with the antibody. A goat anti-mouse FITC antibody was used as the secondary.



Flow Cytometry - Anti-PLK1 antibody [35-206] (ab17056)

Overlay histogram showing HCT116 cells stained with ab17056 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab17056, 1µg/1x10^6 cells) for 30 min at 22ºC. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22ºC. Isotype control antibody (black line) was mouse IgG2b [PLPV219] (ab91366, 2µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HCT116 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

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