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antibody and ELISA experts

BOSTER BIOLOGICAL TECHNOLOGY
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Anti-Cdc25B Antibody Picoband®

Catalog Number: PB9488

About CDC25B

Central to the onset of mitosis in all eukaryotic cells is the CDC2 protein kinase, the activity of which is negatively regulated by phosphorylation and positively activated by dephosphorylation. The latter function is carried out by a specific phosphatase, CDC25. At least 3 human CDC25 genes code for the A, B, and C forms of CDC25. CDC25B is mapped to 20p13. P38 kinase has a critical role in the initiation of a G2 delay after ultraviolet radiation. Inhibition of p38 blocks the rapid initiation of this checkpoint in both human and murine cells after ultraviolet radiation. In vitro, p38 binds and phosphorylates CDC25B at serines 309 and 361, and CDC25C at serine-216; phosphorylation of these residues is required for binding to 14-3-3 proteins. In vivo, inhibition of p38 prevents both phosphorylation of CDC25B at serine-309 and 14-3-3 binding after ultraviolet radiation, and mutation of this site is sufficient to inhibit the checkpoint initiation. Regulation of CDC25B phosphorylation by p38 is a critical event for initiating the G2/M checkpoint after ultraviolet radiation.

Overview

Product Name	Anti-Cdc25B Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-Cdc25B Antibody Picoband® catalog # PB9488. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.
Storage Instructions	Store at -20˚C for one year from date of receipt. After reconstitution, at 4˚C for one month. It can also be aliquotted and stored frozen at -20˚C for six months. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	P30305

Technical Details

Immunogen	E.coli-derived human Cdc25B recombinant protein (Position: M119-L248). Human Cdc25B shares 71% and 68.2% amino acid (aa) sequence identity with mouse and rat Cdc25B, respectively.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins

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Isotype	Rabbit IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.1-0.5ug/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human Immunocytochemistry/Immunofluorescence, 5 ug/ml, Human Flow Cytometry(Fixed), 1-3 ug/1x10^6 cells, Human

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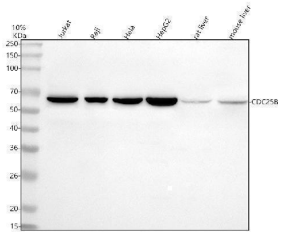
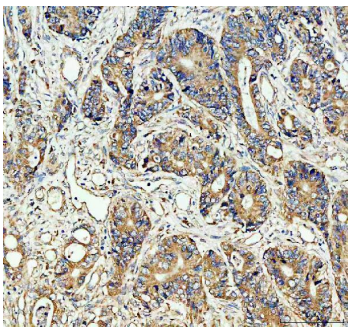
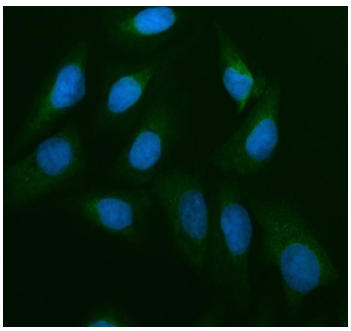
Anti-Cdc25B Antibody Picoband® (PB9488) Images



Western blot analysis of Cdc25B using anti-Cdc25B antibody (PB9488). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human Jurkat whole cell lysates, Lane 2: human Raji whole cell lysates, Lane 3: human Hela whole cell lysates, Lane 4: human HepG2 whole cell lysates, Lane 5: rat liver tissue lysates, Lane 6: mouse liver tissue lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-Cdc25B antigen affinity purified polyclonal antibody (Catalog # PB9488) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for Cdc25B at approximately 65 kDa. The expected band size for Cdc25B is at 65 kDa.



IHC analysis of Cdc25B using anti-Cdc25B antibody (PB9488). Cdc25B was detected in a paraffin-embedded section of human stomach cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-Cdc25B Antibody (PB9488) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



IF analysis of Cdc25B using anti-Cdc25B antibody (PB9488). Cdc25B was detected in an immunocytochemical section of U2OS cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 ug/mL rabbit anti-Cdc25B Antibody (PB9488) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

Flow Cytometry analysis of HepG2 cells using anti-Cdc25B antibody (PB9488). Overlay histogram showing HepG2 cells stained with PB9488 (Blue line). To facilitate intracellular

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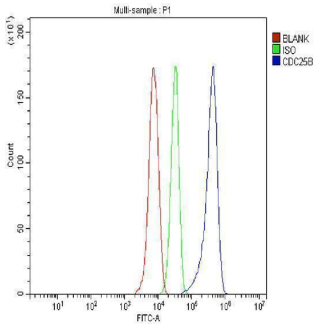

staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-Cdc25B Antibody (PB9488, 1 ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

Anti-Cdc25B Antibody

For Research Use Only. Not for use in diagnostic procedures.