BOSTER BIOLOGICAL TECHNOLOGY
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Anti-Daxx Antibody Picoband®

Catalog Number: PB9550

About DAXX

DAXX (death-domain associated protein) also known as DAP6 (Death-associated protein 6) or BING2, was first discovered through its cytoplasmic interaction with the classical death receptor Fas. Human DAXX encodes a 740-amino acid polypeptide containing a nuclear localization signal. Functional analyses by Yang et al. (1997) demonstrated that Daxx binds to the Fas death domain and enhances Fas-mediated apoptosis. The authors suggested that DAXX and FADD define 2 distinct apoptotic pathways downstream of Fas. The DAXX gene is mapped to human chromosome 6p21.3 by somatic cell hybrid panels and fluorescence in situ hybridization, a region containing the HLA and putative autoimmune disease genes. MSP58 overexpression relieved DAXX-mediated transcriptional repression. Immunoprecipitation and Western blot analysis with DAXX mutants showed that the N terminus of DAXX interacts with the C terminus of DMAP. Transient expression of DAXX or DMAP1 caused repression of glucocorticoid receptor-mediated transcription.

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Overview

Product Name	Anti-Daxx Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-Daxx Antibody Picoband® catalog # PB9550. 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 antibody formulated with stabilizing components, 0.9 mg NaCl, 0.2 mg Na2HPO4, and 0.05 mg NaN3. *This antibody is supplied in a stabilized formulation. Compatibility with conjugation reactions depends on the chemistry of the conjugation method used. For conjugation methods that are not compatible with the stabilizing components present in this formulation, a carrier-free antibody format is required.
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	Q9UER7

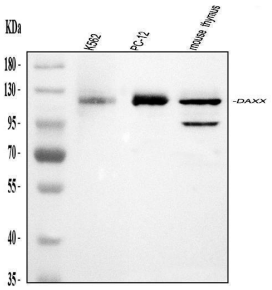
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Technical Details

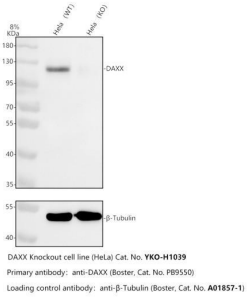
Immunogen	E.coli-derived human Daxx recombinant protein (Position: K56-R345). Human Daxx shares 88.3% and 88.6% amino acid (aa) sequence identity with mouse and rat Daxx, respectively.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western 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
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), 0.5-1ug/ml, Human, Rat Immunocytochemistry , 0.5-1ug/ml, Human Immunocytochemistry/Immunofluorescence, 2ug/ml, Human Flow Cytometry (Fixed), 1-3ug/1x10^6 cells, Human Note: For flow cytometry (FCM), suitable for fixed cells only. Not suitable for FACS or surface flow cytometry using live cells.

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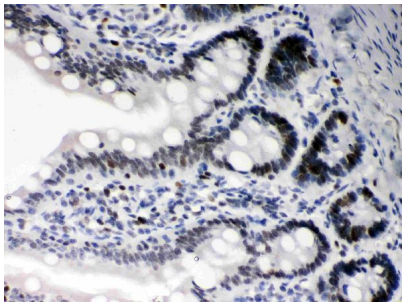
Anti-Daxx Antibody Picoband® (PB9550) Images



Western blot analysis of Daxx using anti-Daxx antibody (PB9550). 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 K562 whole cell lysates, Lane 2: rat PC-12 whole cell lysates, Lane 3: mouse thymus 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-Daxx antigen affinity purified polyclonal antibody (Catalog # PB9550) 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 Daxx at approximately 115 kDa. The expected band size for Daxx is at 81 kDa.

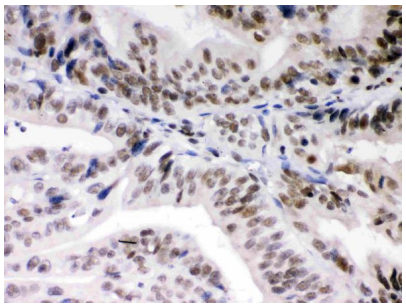


Western blot analysis of DAXX using anti-DAXX antibody (PB9550). Electrophoresis was performed on a 8% SDS-PAGE gel at 80V (Stacking gel) / 120V (Resolving gel) for 2 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human Hela- WT whole cell lysates, Lane 2: human Hela-DAXX KO whole cell 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-DAXX antigen affinity purified polyclonal antibody (PB9550) 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 ECL Plus Western Blotting Substrate (Catalog # AR1196-200) with Tanon 5200 system. A specific band was detected for DAXX at approximately 115 kDa. The expected band size for DAXX is at 81 kDa.

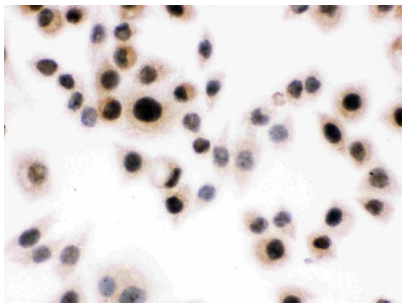


IHC analysis of DAXX using anti-DAXX antibody (PB9550). DAXX was detected in a paraffin-embedded section of rat intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 ug/ml rabbit anti-DAXX Antibody (PB9550) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

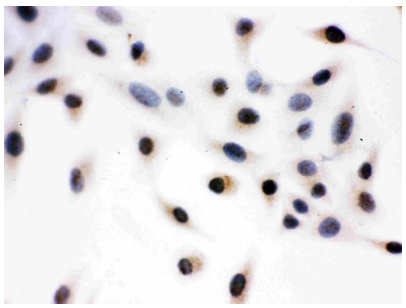
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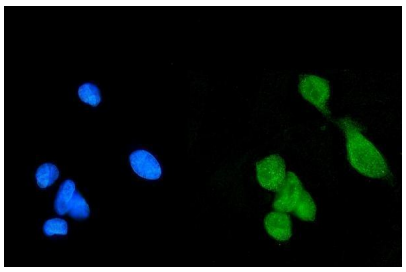
IHC analysis of DAXX using anti-DAXX antibody (PB9550). DAXX was detected in a paraffin-embedded section of human intestinal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 ug/ml rabbit anti-DAXX Antibody (PB9550) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.



IHC analysis of DAXX using anti-DAXX antibody (PB9550). DAXX was detected in immunocytochemical section of SMMC-7721 Cell. 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 1ug/ml rabbit anti-DAXX Antibody (PB9550) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.



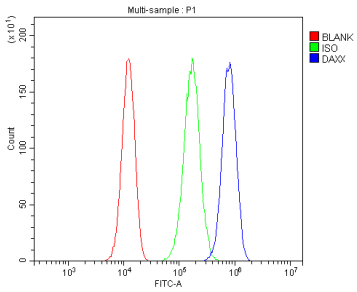
IHC analysis of DAXX using anti-DAXX antibody (PB9550). DAXX was detected in immunocytochemical section of A549 Cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 1ug/ml rabbit anti-DAXX Antibody (PB9550) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.



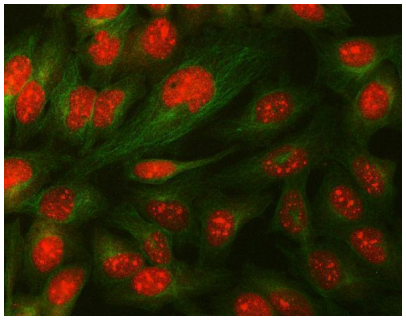
IF analysis of Daxx using anti-Daxx antibody (PB9550). Daxx was detected in immunocytochemical section of U20S cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2ug/mL rabbit anti-Daxx Antibody (PB9550) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:100 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 293T cells using anti-Daxx antibody (PB9550).Overlay histogram showing 293T cells stained with PB9550 (Blue line). To facilitate intracellular 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-Daxx Antibody (PB9550,1ug/1x106 cells) for

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30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



IF analysis of Daxx using anti-Daxx antibody (PB9550) and anti-Tubulin Alpha antibody (M03989-3).Daxx was detected in immunocytochemical section of Hela cell. 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-Daxx Antibody (PB9550) and mouse anti-Tubulin Alpha antibody (M03989-3) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) and DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) were used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

Anti-Daxx Antibody

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