BOSTER BIOLOGICAL TECHNOLOGY
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Anti-XBP1 Antibody Picoband®

Catalog Number: PB9463

About XBP1

XBP1, also known as X-box binding protein 1, is a protein which in humans is encoded by the XBP1 gene. It encodes a transcription factor that regulates MHC class II genes by binding to a promoter element referred to as an X box. This gene product is a bZIP protein, which was also identified as a cellular transcription factor that binds to an enhancer in the promoter of the T cell leukemia virus type 1 promoter. It may increase expression of viral proteins by acting as the DNA binding partner of a viral transactivator. It has been found that upon accumulation of unfolded proteins in the endoplasmic reticulum (ER), the mRNA of this gene is processed to an active form by an unconventional splicing mechanism that is mediated by the endonuclease inositol-requiring enzyme 1 (IRE1). The resulting loss of 26 nt from the spliced mRNA causes a frame-shift and an isoform XBP1 (S), which is the functionally active transcription factor. The isoform encoded by the unspliced mRNA, XBP1 (U), is constitutively expressed, and thought to function as a negative feedback regulator of XBP1 (S), which shuts off transcription of target genes during the recovery phase of ER stress. A pseudogene of XBP1 has been identified and localized to chromosome 5.

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Overview

Product Name	Anti-XBP1 Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-XBP1 Antibody Picoband® catalog # PB9463. 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

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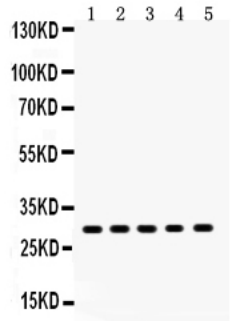
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Technical Details

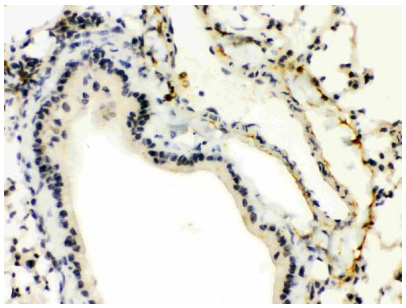
Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human XBP1, identical to the related mouse and rat sequences.
Recommended Detection Systems	Boster provides a series of assays reacted with primary antibodies. Antibody can be supported by chemiluminescence kit EK1002 in WB, supported by SA1022 in 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 Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human, Mouse, Rat 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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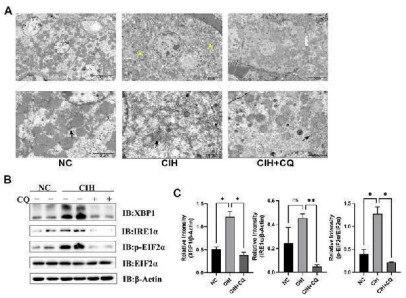
Anti-XBP1 Antibody Picoband® (PB9463) Images



Western blot analysis of XBP using anti-XBP antibody (PB9463). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. Lane 1: MCF-7 Whole Cell Lysate at 40ug, Lane 2: MM231 Whole Cell Lysate at 40ug, Lane 3: MM453 Whole Cell Lysate at 40ug, Lane 4: SKOV Whole Cell Lysate at 40ug, Lane 5: HELA Whole Cell Lysate at 40ug. 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-XBP antigen affinity purified polyclonal antibody (Catalog # PB9463) 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 XBP at approximately 29 kDa. The expected band size for XBP is at 29 kDa.



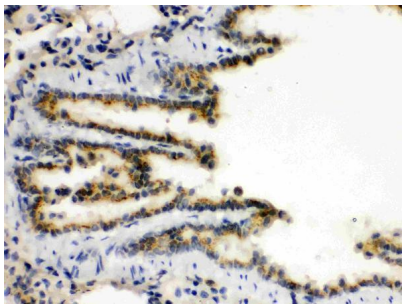
IHC analysis of XBP using anti-XBP antibody (PB9463). XBP was detected in a paraffin-embedded section of mouse lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 ug/ml rabbit anti-XBP Antibody (PB9463) 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.



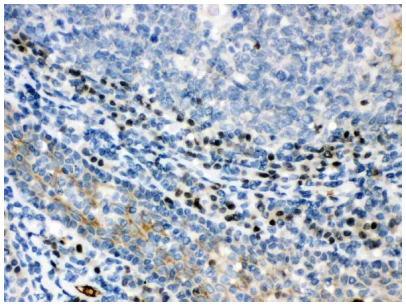
Inhibition of autophagic flux alleviated hepatocyte ER stress and LD accumulation. (A) Changes in ER morphology and LD accumulation in different groups were assessed. (B) Hepatic XBP1, IRE1alpha, and EIF2alpha protein levels were compared among groups. (C) XBP1 and IRE1alpha protein levels were normalized based on beta-Actin levels, and the p-EIF2alpha/EIF2alpha ratio was compared among groups. (Yellow arrow: LDs; black arrow: ER; * $p < 0.05$, ** $p < 0.01$, ns: no statistical difference).Index in PubMed under a CC BY license. PMID: 35982710

IHC analysis of XBP using anti-XBP antibody (PB9463). XBP was detected in a paraffin-embedded section of rat lung 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-XBP Antibody (PB9463) overnight at 4°C. Biotinylated goat anti-

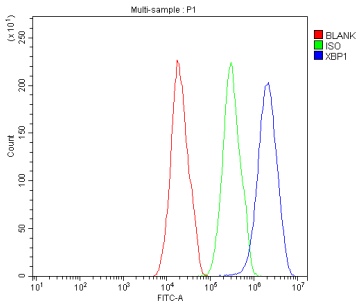
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rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.



IHC analysis of XBP using anti-XBP antibody (PB9463). XBP was detected in a paraffin-embedded section of human tonsil 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-XBP Antibody (PB9463) 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.



Flow Cytometry analysis of HepG2 cells using anti-XBP1 antibody (PB9463). Overlay histogram showing HepG2 cells stained with PB9463 (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-XBP1 Antibody (PB9463, 1ug/1x106 cells) for 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.

2 Publications Citing This Product

1. PubMed ID: 10.1002/tox.20724, Excessive fluoride induces endoplasmic reticulum stress and interferes enamel proteinases secretion

2. PubMed ID: -, Wang,X.,Xu,X.,Yang,Y. P.,Xin,X.,Li,Z.,Wang,Q. ... Zhang,L.(2021).Dihydromyricetin alleviates endothelial inflammatory response through IRE1alpha/NF-kappaB signaling pathway in sepsis.Archives of Medical Science.https://doi.org/10.5114/aoms/132293

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