Product datasheet

Anti-XBP-1U/XBP1 specific Antibody

Catalog Number: *PB9463*

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Basic Information

Product Name	Anti-XBP-1U/XBP1 specific Antibody
Gene Name	XBP1
Source	Rabbit
Isotype	IgG
Species Reactivity	human, mouse, rat
Tested Application	WB, IHC, IHC-F, ICC, ICC/IF, FCM
Contents	500 ug/ml antibody with PBS， 0.02% NaN3 , 1 mg BSA and 50% glycerol.
Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human XBP1 (59-85 aa RKRQRLTHLSPEEKALRRKLKNRVAAQ), identical to the related mouse and rat sequences.
concentration	500 ug/ml
Purification	Immunogen affinity purified.
Observed MW	29KD
Dilution Ratios	Western blot(WB): 1:500-2000 Immunohistochemistry in paraffin section (IHC): 1:50-400 Immunohistochemistry in frozen section (IHC-F): 1:50-400 Immunocytochemistry: 1:50-400 Flow cytometry (FCM): 1-3 μg/1x10^6 cells Immunocytochemistry/Immunofluorescence (ICC/IF): 1:50-400 (Boiling the paraffin sections in 10mM citrate buffer,pH6.0,or PH8.0 EDTA repair liquid for 20 mins is required for the staining of formalin/paraffin sections.) Optimal working dilutions must be determined by end user.

Storage

12 months from date of receipt，-20℃ as supplied. 6 months 2 to 8℃ after reconstitution. Avoid repeated freezing and thawing.

Background Information

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

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been identified and localized to chromosome 5.

Reference

Anti-XBP-1U/XBP1 specific Antibody被引用在1文献中。

Selected Validation Data

Figure 1. Western blot analysis of XBP1 using anti-XBP1 antibody (PB9463). 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 50ug of sample under reducing conditions. Lane 1: MCF-7 Whole Cell LysateLane 2: MM231 Whole Cell LysateLane 3: MM453 Whole Cell LysateLane 4: SKOV Whole Cell LysateLane 5: HELA Whole Cell Lysate After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA 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-XBP1 antigen affinity purified polyclonal antibody (Catalog # PB9463) at 0.5 µg/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 1:10000 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 XBP1 at approximately 29KD. The expected band size for XBP1 is at 29KD.

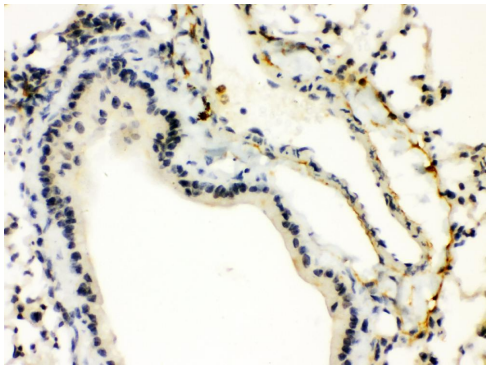

Figure 2. IHC analysis of XBP using anti-XBP antibody (PB9463).XBP was detected in paraffin-embedded section of mouse lung tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1µg/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.

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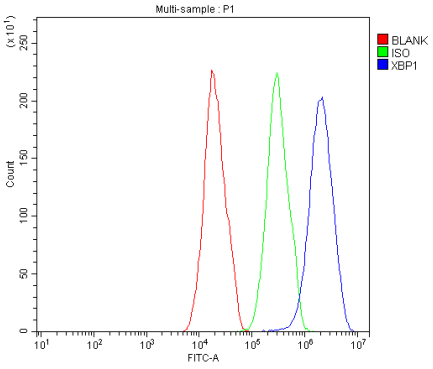

Figure 5. Flow Cytometry analysis of HepG2 cells using anti-XBP1 antibody (PB9463).
Overlay histogram showing HepG2 cells stained with PB9463 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti- XBP1 Antibody (PB9463, 1µg/1x10^6 cells) for 30 min at 20°C. DyLight488 conjugated goat anti-rabbit IgG (BA1127, 5-10µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

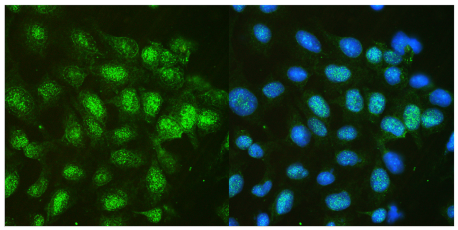

Figure 6. IF analysis of XBP1 using anti-XBP1 antibody (PB9463). XBP1 was detected in immunocytochemical section of U2OS cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) rabbit anti-XBP1 Antibody (PB9463). DyLight 488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.