Anti-YB1 Antibody [ST0432]

ET1609-10

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 36 kDa
Clone number:	ST0432

Description: Y-box binding protein YB-1 (also known as CCAAT-binding transcription factor, enhancer factor I subunit A and DNA-binding protein B) belongs to a family of multifunctional proteins which regulate both transcription and translation. Y-box proteins interact with a wide variety of nucleic acid structures to act as transcription factors and mRNA masking proteins. The modular structure of Y-box proteins includes a highly conserved N-terminal cold-shock domain (CSD, equivalent to the bacterial cold-shock proteins) and four basic C-terminal domains containing Arginine clusters and aromatic residues. YB-1 plays a role in cell proliferation as an activator of growth-associated gene expression. YB-1 is also a repressor of the cell death-associated gene FAS. YB-1 may play an important role in controlling cell survival by regulating the expression of cell growth-associated and death-associated genes.

Immunogen: Synthetic peptide within Human YB1 aa 275-324 / 324.

Positive control: HeLa cell lysate, Jurkat cell lysate, PC-12, human kidney tissue, mouse colon tissue, mouse stomach tissue, human prostate carcinoma tissue, Hela.

Subcellular location: Cytoplasm, Nucleus, Cytoplasmic granule, Secreted.

Database links: SwissProt: P67809 Human | P62960 Mouse | P62961 Rat

Recommended Dilutions:

WB	1:1,000-1:5,000
IF-Cell	1:100-1:500
IF-Tissue	1:100-1:500
IHC-P	1:50-1:200
FC	1:50-1:100
IP	Use at an assay dependent concentration.

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

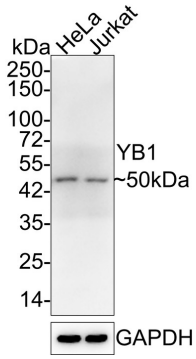

Fig1: Western blot analysis of YB1 on different lysates with Rabbit anti-YB1 antibody (ET1609-10) at 1/1,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: Jurkat cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 36 kDa
Observed band size: 50 kDa

Exposure time: 2 minutes 18 seconds;
4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1609-10) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

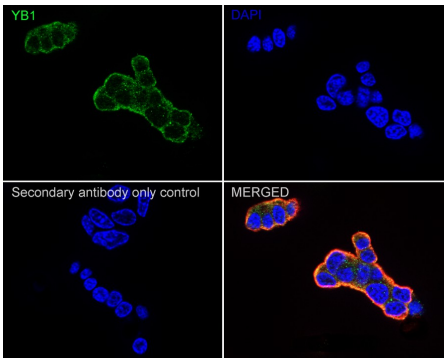

Fig2: Immunocytochemistry analysis of PC-12 cells labeling YB1 with Rabbit anti-YB1 antibody (ET1609-10) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-YB1 antibody (ET1609-10) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

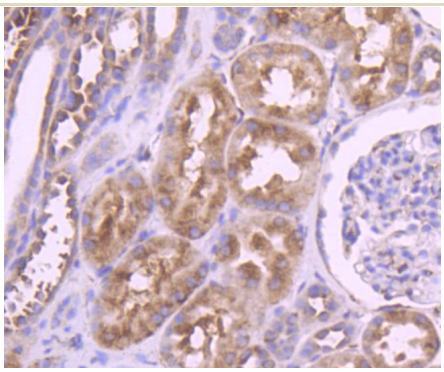

Fig3: Immunohistochemical analysis of paraffin-embedded human kidney tissue using anti-YB1 antibody at 1/200. Counter stained with hematoxylin.

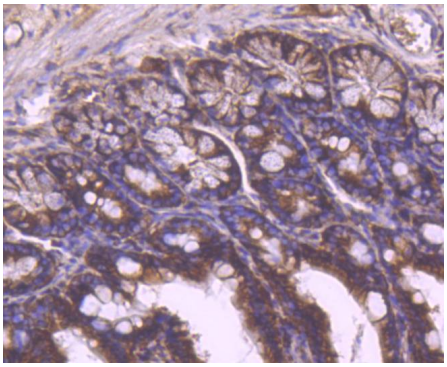

Fig4: Immunohistochemical analysis of paraffin-embedded mouse colon tissue using anti-YB1 antibody at 1/200. Counter stained with hematoxylin.

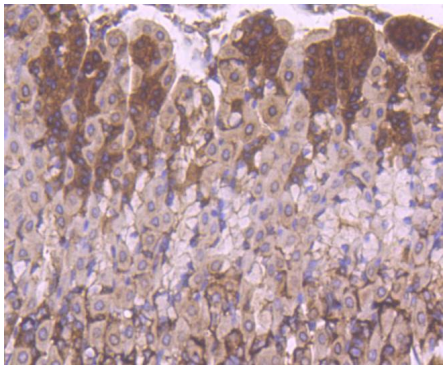

Fig5: Immunohistochemical analysis of paraffin-embedded mouse stomach tissue using anti-YB1 antibody at 1/200. Counter stained with hematoxylin.

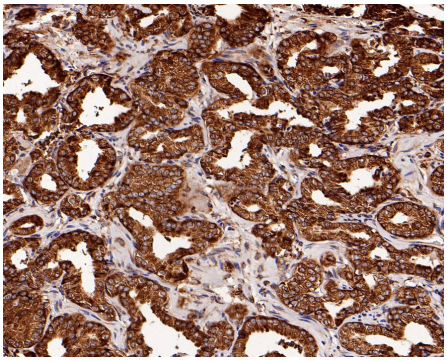

Fig6: Immunohistochemical analysis of paraffin-embedded human prostate carcinoma tissue with Rabbit anti-YB1 antibody (ET1609-10) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1609-10) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

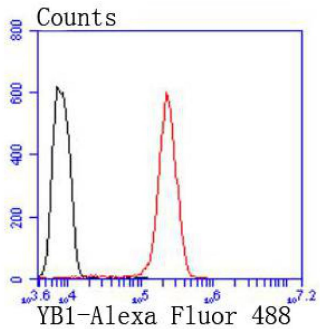

Fig7: Flow cytometric analysis of Hela cells with YB1 antibody at 1/50 dilution (red) compared with an unlabelled control (cells without incubation with primary antibody; black). Alexa Fluor 488-conjugated goat anti rabbit IgG was used as the secondary antibody.

Background References

1. Satkunanathan S et al. Establishment of a novel cell line for the enhanced production of recombinant adeno-associated virus vectors for gene therapy. Hum Gene Ther 25:929-41 (2014).
2. Imada K et al. Mutual regulation between Raf/MEK/ERK signaling and Y-box-binding protein-1 promotes prostate cancer progression. Clin Cancer Res 19:4638-50 (2013).