

Anti-BTK Antibody [SN69-07]

ET1611-4



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IHC-P, IP
Molecular Wt:	Predicted band size: 76 kDa
Clone number:	SN69-07

Description: The Tec family of non-receptor tyrosine kinases is composed of six proteins designated Tec, Emt (also known as Itk or Tsk), Btk (previously known as Atk, BPK or Emb), Bmx, Txk (also known as Rlk) and Dsrc28C. All members of the family contain SH3 and SH2 domains and, with the exception of Txk and Dsrc28C, also contain a pleckstrin homology (PH) and a Tec homology (TH) domain in their amino termini. Four alternatively spliced forms of Tec are found to be expressed broadly in cells of hematopoietic lineage and hepatocytes. The Emt gene product associates with CD28 and becomes activated subsequent to CD28 ligation. Btk is necessary for proper B cell development, and mutations in the gene encoding Btk have been associated with families suffering from X-linked agammaglobulinemia, also referred to as Bruton's disease. The Bmx protein shares a high degree of homology with Btk and seems to be expressed at highest levels in the heart. Txk expression is T cell-specific, while expression of the Drosophila Tec homolog, Dsrc28C, is developmentally regulated.

Immunogen: Synthetic peptide within Human BTK aa 610-659 / 659.

Positive control: Ramos cell lysate, Raji cell lysate, THP-1 cell lysate, Daudi cell lysate, RAW264.7 cell lysate, human tonsil tissue.

Subcellular location: Cell membrane, Cytoplasm, Membrane, Nucleus.

Database links: SwissProt: Q06187 Human | P35991 Mouse

Recommended Dilutions:

WB	1:5,000
IHC-P	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: Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.

Purity: Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders: 0086-571-88062880

Technical: 0086-571-89986345

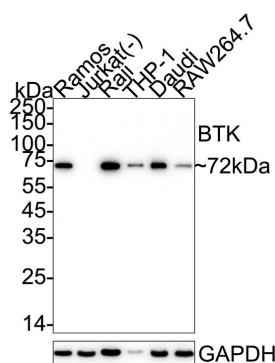
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Applications: WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

Fig1: Western blot analysis of BTK on different lysates with Rabbit anti-BTK antibody (ET1611-4) at 1/5,000 dilution.



Lane 1: Ramos cell lysate

Lane 2: Jurkat cell lysate (negative)

Lane 3: Raji cell lysate

Lane 4: THP-1 cell lysate

Lane 5: Daudi cell lysate

Lane 6: RAW264.7 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 76 kDa

Observed band size: 72 kDa

Exposure time: 3 minutes; ECL: K1801;

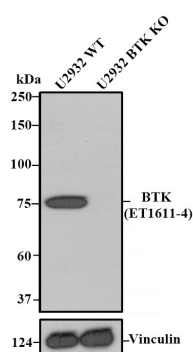
4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1611-4) at 1/5,000 dilution was used in primary antibody dilution (K1803) at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

Fig2: All lanes: Western blot analysis of BTK with anti-BTK antibody [SN69-07] (ET1611-4) at 1/1,000 dilution.

Lane 1: Wild-type U2932 whole cell lysate.

Lane 2: BTK knockout U2932 whole cell lysate.



ET1611-4 was shown to specifically react with BTK in wild-type U2932 cells. No band was observed when BTK knockout sample was tested. Wild-type and BTK knockout samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary Anti-BTK antibody (ET1611-4, 1/1,000) and Anti-Vinculin antibody (ET1705-94, 1/5,000) were used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG H&L (HRP) Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

Cell lysate was provided by Ubigen Biosciences (Ubigen Biosciences Co., Ltd., Guangzhou, China).

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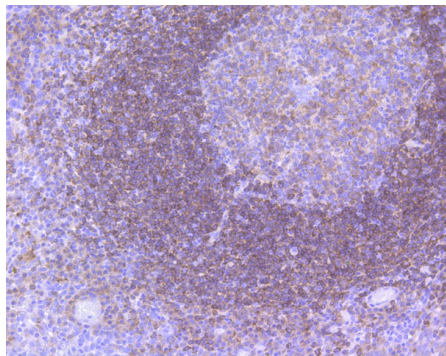


Fig3: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-BTK antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1611-4, 1/50) for 30 minutes 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.

Note: All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

Background References

1. Landon AL et al. MNKs act as a regulatory switch for eIF4E1 and eIF4E3 driven mRNA translation in DLBCL. *Nat Commun* 5:5413 (2014).
2. Bogusz AM et al. Quantitative Immunofluorescence Reveals the Signature of Active B-cell Receptor Signaling in Diffuse Large B-cell Lymphoma. *Clin Cancer Res* 18:6122-35 (2012).

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