abcam

Product datasheet

Anti-BTK (phospho Y223) antibody [EP420Y] ab68217

Recombinant RabMAb

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Overview

Product name	Anti-BTK (phospho Y223) antibody [EP420Y]
Description	Rabbit monoclonal [EP420Y] to BTK (phospho Y223)
Host species	Rabbit
Tested applications	**Suitable for:** WB, IP, Dot blot **Unsuitable for:** ICC/IF or IHC-P
Species reactivity	**Reacts with:** Human **Predicted to work with:** Mouse, Rat
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. (Peptide available as **ab214589**)
Positive control	WB: K562 and Ramos cell lysates treated with pervanadate. IP: Ramos cell lysates treated with pervanadate. Dot Blot: BTK (phospho Y223) phospho peptide.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. **We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.**

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer	pH: 7.2

Preservative: 0.01% Sodium azide
Constituents: 50% Glycerol (glycerin, glycerine), 0.05% BSA

Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP420Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab68217 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/2000 - 1/10000. Detects a band of approximately 76 kDa (predicted molecular weight: 76 kDa).Can be blocked with **BTK peptide (ab214589)**. **For unpurified, use 1/10000 - 1/50000.**
IP		1/20. **For unpurified, use 1/40.**
Dot blot		Use at an assay dependent concentration.

Application notes Is unsuitable for ICC/IF or IHC-P.

Target

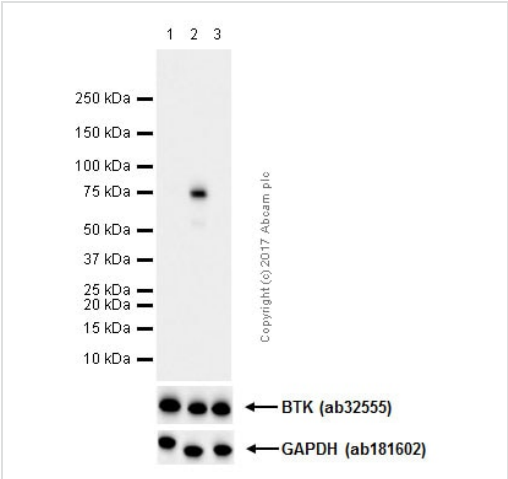
Function Plays a crucial role in B-cell ontogeny. Transiently phosphorylates GTF2I on tyrosine residues in response to B-cell receptor cross-linking. Required for the formation of functional ARID3A DNA-binding complexes.

Involvement in disease Defects in BTK are the cause of X-linked agammaglobulinemia (XLA) [MIM:300755]; also known as X-linked agammaglobulinemia type 1 (AGMX1) or immunodeficiency type 1 (IMD1). XLA is a humoral immunodeficiency disease which results in developmental defects in the maturation pathway of B-cells. Affected boys have normal levels of pre-B-cells in their bone marrow but virtually no circulating mature B-lymphocytes. This results in a lack of immunoglobulins of all classes and leads to recurrent bacterial infections like otitis, conjunctivitis, dermatitis, sinusitis in the first few years of life, or even some patients present overwhelming sepsis or meningitis, resulting in death in a few hours. Treatment in most cases is by infusion of intravenous immunoglobulin.
Defects in BTK may be the cause of X-linked hypogammaglobulinemia and isolated growth hormone deficiency (XLA-IGHD) [MIM:307200]; also known as agammaglobulinemia and isolated growth hormone deficiency or Fleisher syndrome or isolated growth hormone deficiency type 3 (IGHD3). In rare cases XLA is inherited together with isolated growth hormone deficiency (IGHD).

Sequence similarities Belongs to the protein kinase superfamily. Tyr protein kinase family. TEC subfamily.
Contains 1 Btk-type zinc finger.
Contains 1 PH domain.
Contains 1 protein kinase domain.
Contains 1 SH2 domain.
Contains 1 SH3 domain.

Post-translational modifications	Autophosphorylated on Tyr-223 and Tyr-551. Phosphorylation of Tyr-223 may create a docking site for a SH2 containing protein.
Cellular localization	Cytoplasm. Membrane. Nucleus.

Images



Western blot - Anti-BTK (phospho Y223) antibody [EP420Y] (ab68217)

All lanes : Anti-BTK (phospho Y223) antibody [EP420Y] (ab68217) at 1/1000 dilution

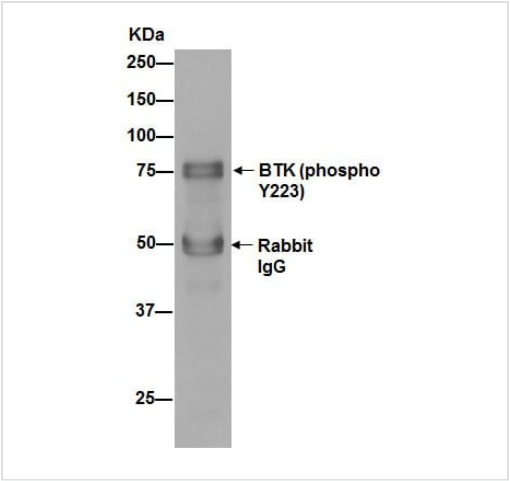
Lane 1 : Ramos (human Burkitt's lymphoma B lymphocyte) whole cell lysate
Lane 2 : Ramos treated with 1mM pervanadate for 30 min whole cell lysate
Lane 3 : Ramos treated with 1mM pervanadate for 30 min whole cell lysate. Then the membrane was incubated with alkaline phosphatase.

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 76 kDa
Observed band size: 76 kDa

Blocking and dilution buffer: 5% NFDM/TBST.

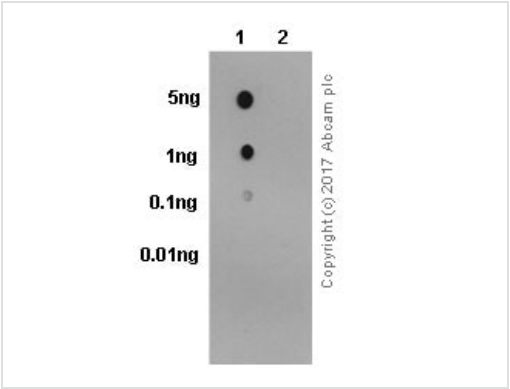


Immunoprecipitation - Anti-BTK (phospho Y223) antibody [EP420Y] (ab68217)

ab68217 (purified) at 1/50 immunoprecipitating BTK (phospho Y223) in Ramos cells (Lane 1) treated with 1 mM Pervanadate for 30 min. For western blotting, a HRP-conjugated anti-rabbit IgG (H+L) was used as the secondary antibody (1/1000).

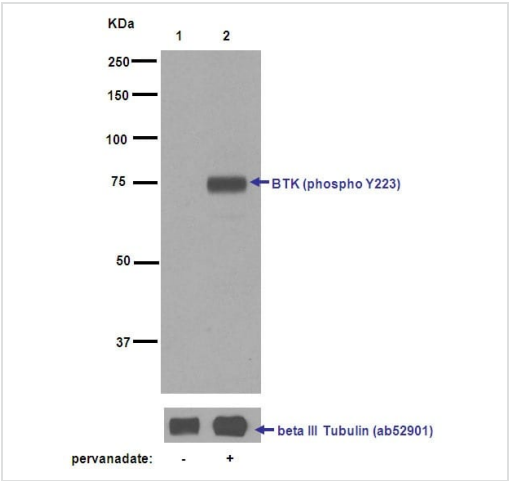
Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Dot Blot - Anti-BTK (phospho Y223) antibody [EP420Y] (ab68217)

Dot blot analysis of BTK (phospho Y223) phospho peptide (Lane 1) and BTK phospho peptide (Lane 2) labeling BTK (phospho Y223) with ab68217 at a dilution of 1/1000. A Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (**ab97051**) was used as the secondary antibody at a dilution of 1/20,000. Blocking buffer: 5% NFDM/TBST. Dilution buffer: 5% NFDM /TBST.



Western blot - Anti-BTK (phospho Y223) antibody [EP420Y] (ab68217)

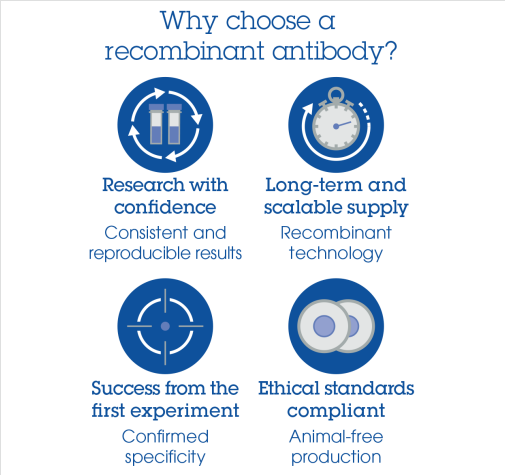
All lanes : Anti-BTK (phospho Y223) antibody [EP420Y] (ab68217) at 1/10000 dilution (purified)

Lane 1 : Untreated K562 cell lysate
Lane 2 : K562 cells treated with pervanadate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 76 kDa
Observed band size: 76 kDa



Anti-BTK (phospho Y223) antibody [EP420Y]

(ab68217)

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