abcam

Product datasheet

Anti-ZAP70 (phospho Y292) antibody [EPR1073] ab76501

Recombinant RabMAb

★★★★ 1 Abreviews

5 Images

Overview

Product name Anti-ZAP70 (phospho Y292) antibody [EPR1073]

Description Rabbit monoclonal [EPR1073] to ZAP70 (phospho Y292)

Host species Rahhit

Tested applications Suitable for: Flow Cyt (Intra), WB, ICC/IF, Dot blot

Unsuitable for: IHC-P or IP

Species reactivity Reacts with: Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Jurkat whole cell lysate (ab7899) treated with pervanadate. ICC/IF: Untreated and

pervanadate treated Jurkat cells. Flow Cyt (intra): Untreated and pervanadate treated Jurkat cells.

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**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

Storage buffer pH: 7.20

Preservative: 0.05% Sodium azide

Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue

culture supernatant

Purity Protein A purified

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ClonalityMonoclonalClone numberEPR1073

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab76501 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
Flow Cyt (Intra)		1/20. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB	★★☆☆☆ (1)	1/5000 - 1/10000. Predicted molecular weight: 70 kDa.
ICC/IF		1/250 - 1/500.
Dot blot		1/1000.

Application notes Is unsuitable for IHC-P or IP.

Target

Function Plays a role in T-cell development and lymphocyte activation. Essential for TCR-mediated IL-2

production. Isoform 1 induces TCR-mediated signal transduction, isoform 2 does not.

Tissue specificity Expressed in T- and natural killer cells.

Involvement in disease Defects in ZAP70 are the cause of selective T-cell defect (STD) [MIM:176947]. STD is an

autosomal recessive form of severe combined immunodeficiency characterized by a selective

absence of CD8-type T-cells.

Sequence similaritiesBelongs to the protein kinase superfamily. Tyr protein kinase family. SYK/ZAP-70 subfamily.

Contains 1 protein kinase domain.

Contains 2 SH2 domains.

Domain The SH2 domains bind to the phosphorylated tyrosine-based activation motif (TAM) of CD3Z and

the non-canonical phosphorylated tyrosine-based activation motif (TAM) of RHOH.

Post-translational

modifications

Phosphorylated on tyrosine residues upon T-cell antigen receptor (TCR) stimulation. Tyr-319

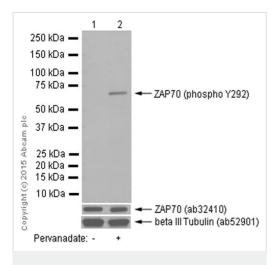
phosphorylation is essential for full activity.

Cytoplasm. Cell membrane. After antigen stimulation, isoform 1 concentrates at the

immunological synapse and isoform 2 remains cytoplasmic. Co-localizes together with RHOH in the immunological synapse. RHOH is required for its proper localization to the cell membrane and

cytoskeleton fractions in the thymocytes.

Images



Western blot - Anti-ZAP70 (phospho Y292) antibody [EPR1073] (ab76501)

All lanes : Anti-ZAP70 (phospho Y292) antibody [EPR1073] (ab76501) at 1/5000 dilution

Lane 1 : Untreated Jurkat (human acute T cell leukemia) whole cell lysates

Lane 2: Jurkat (human acute T cell leukemia) treated with pervanadate whole cell lysates

Lysates/proteins at 10 µg per lane.

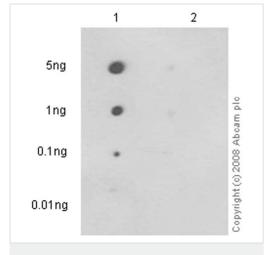
Secondary

All lanes : Peroxidase-conjugated goat anti-rabbit lgG, (H+L) at 1/1000 dilution

Predicted band size: 70 kDa **Observed band size:** 70 kDa

Exposure time: 3 minutes

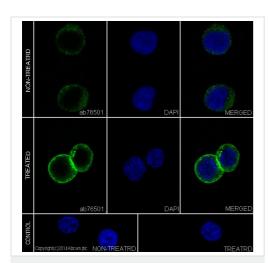
Blocking and dilution buffer: 5% NFDM/TBST.



Dot Blot - Anti-ZAP70 (phospho Y292) antibody [EPR1073] (ab76501)

Dot blot analysis of varying amounts of ZAP70 (pY292) phospho peptide (Lane 1) and ZAP70 non-phospho peptide (Lane 2) using ab76501 at a dilution of 1/1000. A peroxidase-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/2500).

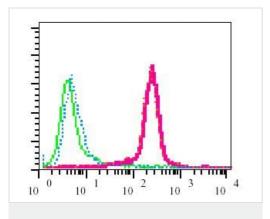
Exposure time: 3 minutes.



Immunocytochemistry/ Immunofluorescence - Anti-ZAP70 (phospho Y292) antibody [EPR1073] (ab76501)

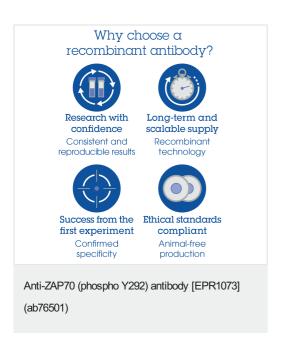
Immunocytochemistry/Immunofluorescence analysis of untreated and pervanadate treated Jurkat (human acute T cell leukemia) cells labelling ZAP70 (phospho Y292) with ab76501 at a dilution of 1/250. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Treated samples were starved overnight followed by incubation with 1mM pervanadate for 30 minutes. ab150077, an Alexa Fluor 488-conjugated goat antirabbit IgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. Negative control using secondary antibody only.

Image shows increased cytoplasmic staining after pervanadate treatment.



Flow Cytometry (Intracellular) - Anti-ZAP70 (phospho Y292) antibody [EPR1073] (ab76501)

Intracellular flow cytometric analysis of permeabilized Jurkat cells, untreated (green), pervanadate treated (red) stained with ab76501, at 1/20 dilution (blue) and pervanadate-treated Jurkat cells using the same antibody preincubated with phospho- ZAP70 (Tyr 292) peptide (blue) or non-phospho- ZAP70 (Tyr 292)).



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