abcam

Product datasheet

Anti-ZAP70 (phospho Y292) antibody [EPR1073] - BSA and Azide free ab247483

Recombinant

RabMAb

5 Images

Overview

Product name Anti-ZAP70 (phospho Y292) antibody [EPR1073] - BSA and Azide free

Description Rabbit monoclonal [EPR1073] to ZAP70 (phospho Y292) - BSA and Azide free

Host species Rabbit

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

Unsuitable for: IHC-P or IP

Species reactivity Reacts with: Human

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

General notes ab247483 is the carrier-free version of <u>ab76501</u>.

Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our <u>conjugation kits</u> for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar[®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.

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

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these species. Please contact us for more information.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Affinity purified
Clonality Monoclonal
Clone number EPR1073

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab247483 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)		Use at an assay dependent concentration.
Dot blot		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 70 kDa.
ICC/IF		Use at an assay dependent concentration.

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 diseaseDefects 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 similarities Belongs 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

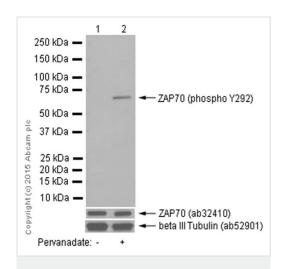
Cellular localization

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] - BSA and Azide free (ab247483) **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

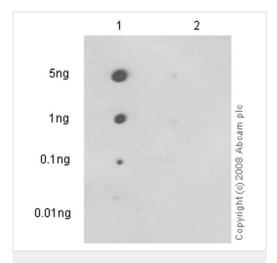
All lanes : Peroxidase-conjugated goat anti-rabbit lgG, (H+L) at 1/1000 dilution

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

Exposure time: 3 minutes

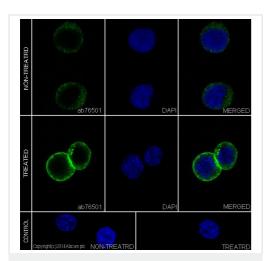
This data was developed using <u>ab76501</u>, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.



Dot Blot - Anti-ZAP70 (phospho Y292) antibody [EPR1073] - BSA and Azide free (ab247483)

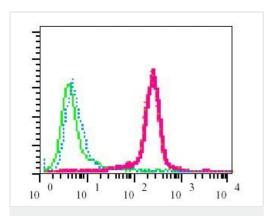
This data was developed using <u>ab76501</u>, the same antibody clone in a different buffer formulation. Dot blot analysis of varying amounts of ZAP70 (pY292) phospho peptide (Lane 1) and ZAP70 non-phospho peptide (Lane 2) using <u>ab76501</u> at a dilution of 1/1000. A peroxidase-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/2500). Exposure time: 3 minutes.



Immunocytochemistry/ Immunofluorescence - Anti-ZAP70 (phospho Y292) antibody [EPR1073] - BSA and Azide free (ab247483)

This data was developed using <u>ab76501</u>, the same antibody clone in a different buffer

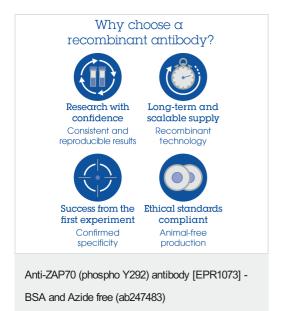
formulation.lmmunocytochemistry/Immunofluorescence analysis of untreated and pervanadate treated Jurkat (human acute T cell leukemia) cells labelling ZAP70 (phospho Y292) with <u>ab76501</u> at a dilution of 1/250. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Treated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



Flow Cytometry (Intracellular) - Anti-ZAP70 (phospho Y292) antibody [EPR1073] - BSA and Azide free (ab247483)

This data was developed using <u>ab76501</u>, the same antibody clone in a different buffer formulation.

Intracellular flow cytometric analysis of permeabilized Jurkat cells, untreated (green), pervanadate treated (red) stained with <u>ab76501</u>, 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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