abcam

Product datasheet

Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] - BSA and Azide free ab238955

Recombinant

RabMAb

5 Images

Overview

Product name Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] - BSA and Azide free

Description Rabbit monoclonal [EP776(2)Y] to CD3 zeta (phospho Y83) - BSA and Azide free

Host species Rabbit

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

Unsuitable for: IHC-P

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). IP: Jurkat. ICC: Jurkat cells. Flow Cyt (intra): Jurkat cells.

General notes ab238955 is the carrier-free version of <u>ab68236</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 **conjugation kits** 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**.

1

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. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

ClonalityMonoclonalClone numberEP776(2)Y

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab238955 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.
WB		Use at an assay dependent concentration. Detects a band of approximately 18-22 kDa (predicted molecular weight: 18 kDa).
Dot blot		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.

Application notes Is unsuitable for IHC-P.

Target

Function Probable role in assembly and expression of the TCR complex as well as signal transduction

upon antigen triggering.

Involvement in disease Defects in CD247 are the cause of immunodeficiency due to defect in CD3-zeta (CD3ZID)

 $[MIM:610163]. \ An immunological \ deficiency \ characterized \ by \ T-cells \ impaired \ immune \ response$

to alloantigens, tetanus toxoid and mitogens.

Sequence similarities Belongs to the CD3Z/FCER1G family.

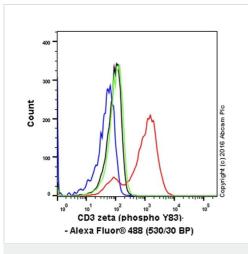
Contains 3 ITAM domains.

Domain The ITAM domains mediate interaction with SHB.

Cellular localization

Membrane.

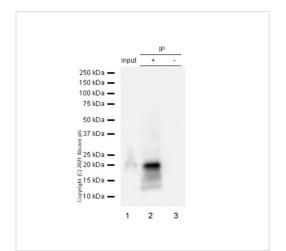
Images



Flow Cytometry (Intracellular) - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] - BSA and Azide free (ab238955)

Flow Cytometry analysis of Jurkat (human acute T cell leukemia) treated (Red)/untreated (Green) with 1mM pervanadate for 4 hours with purified ab68236 at 1/250 dilution. The secondary antibody was Goat anti rabbit lgG (Alexa Fluor[®] 488) at 1/2000 dilution. A Rabbit monoclonal lgG (Black) was used as the isotype control and cells without incubation with primary antibody and secondary antibody (Blue) were used as unlabeled control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab68236).



Immunoprecipitation - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] - BSA and Azide free (ab238955)

This data was developed using <u>ab68236</u>, the same antibody clone in a different buffer formulation.

CD3 zeta was immunoprecipitated from 0.35 mg Jurkat (Human T cell leukemia T lymphocyte) treated with pervandate (50mM 5min) whole cell lysate 10 μ g with <u>ab68236</u> at 1/30 dilution (2 μ g) .

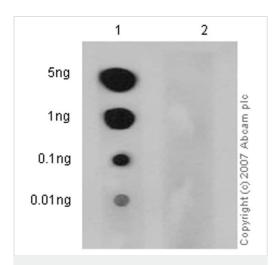
VeriBlot for IP Detection Reagent (HRP)(<u>ab131366</u>) was used at 1/5000 dilution.

Lane 1: Jurkat (Human T cell leukemia T lymphocyte) treated with pervandate (50mM 5min) whole cell lysate 10 µg

Lane 2: abab68236 IP in Jurkat treated with pervandate (50mM 5min) whole cell lysate

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of <u>ab68236</u> in Jurkat treated with pervandate (50mM 5min) whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.



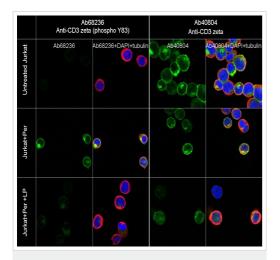
Dot Blot - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] - BSA and Azide free (ab238955)

Dot blot analysis of CD3 zeta (pY83) phospho peptide (lane 1) and CD3 zeta non-phospho peptide (lane 2) labelling CD3 zeta (phospho Y83) with **ab68236** at a dilution of 1/1000. A peroxidase-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/2500).

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab68236).



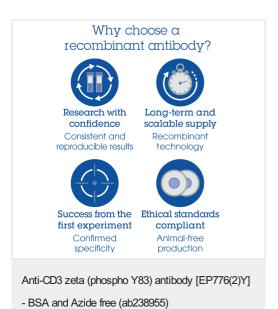
Immunocytochemistry/ Immunofluorescence - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] -BSA and Azide free (ab238955)

Immunocytochemistry/Immunofluorescence analysis of Jurkat cells (untreated, Per treated and Per+LP treated) labelling CD3 zeta (phospho Y83) with <u>ab68236</u> (left) and CD3 zeta with <u>ab40804</u> (right) both at a dilution of 1/200. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. <u>ab150077</u>, an Alexa Fluor[®] 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. <u>ab7291</u>, a mouse anti-tubulin (1/1000) and <u>ab150120</u>, an Alexa Fluor[®] 594-conjugated goat anti-mouse IgG (1/1000) were also used.

The image shows increased cytoplamic staining after Pervanadate (1 mM, 30 min) treatment on Jurkat cells. The LP treatment decreased the cytoplasmic staining caused by Pervanadate.

ab40804 was used as a Pan control for **ab68236**. The results showed cytoplamic staining on untreated, pervanadate (1 mM, 30 min) treated and Per+LP treated Jurkat cells.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab68236).



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