abcam

Product datasheet

Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] ab68236



重组 RabMAb

14 References 7 图像

概述

产品名称 Anti-CD3 zeta (phospho Y83)抗体[EP776(2)Y]

描述 兔单克隆抗体[EP776(2)Y] to CD3 zeta (phospho Y83)

宿主 Rabbit

经测试应用 适用于: Flow Cyt (Intra), ICC/IF, Dot blot, WB, IP

不适用于: IHC-P

种属反应性 与反应: Human

免疫原 Synthetic peptide within Human CD3 zeta aa 50-150 (phospho Y83). The exact sequence is

proprietary.

Database link: P20963

阳性对照 WB: Jurkat whole cell lysate (ab7899). IP: Jurkat. ICC: Jurkat cells. Flow Cyt (intra): Jurkat cells.

常规说明 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 $\mathsf{RabMAb}^{\texttt{®}}$ 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.

性能

形式 Liquid

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

Avoid freeze / thaw cycle.

存储溶液 pH: 7.2

Preservative: 0.05% Sodium azide

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

纯度 Protein A purified

1

克隆 单克隆

克隆编号 EP776(2)Y

同种型 IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab68236于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		Use at an assay dependent concentration.
ICC/IF		1/100 - 1/250.
Dot blot		Use at an assay dependent concentration.
WB		1/5000 - 1/10000. Detects a band of approximately 18-22 kDa (predicted molecular weight: 18 kDa).
IP		1/50.

应用说明 Is unsuitable for IHC-P.

靶标

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

upon antigen triggering.

疾病相关 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.

序列相似性 Belongs to the CD3Z/FCER1G family.

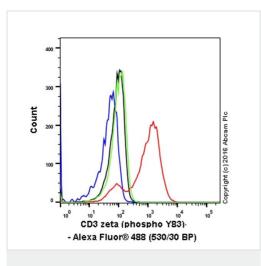
Contains 3 ITAM domains.

结构域 The ITAM domains mediate interaction with SHB.

翻译后修饰 Phosphorylated on Tyr residues after T-cell receptor triggering.

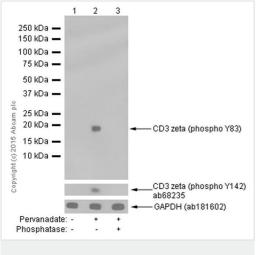
细胞定位 Membrane.

图片



Flow Cytometry (Intracellular) - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] (ab68236)

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.



Western blot - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] (ab68236)

All lanes : Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] (ab68236) at 1/2000 dilution

Lane 1: Untreated Jurkat cells whole cell lysates

Lane 2: Jurkat cells were treated with 50mM Pervanadate for 5 minutes whole cell lysates

Lane 3: Jurkat cells were treated with 50mM Pervanadate for 5 minutes whole cell lysates. Then the membrane was incubated with Alkaline phosphatase.

Lysates/proteins at 10 µg per lane.

Secondary

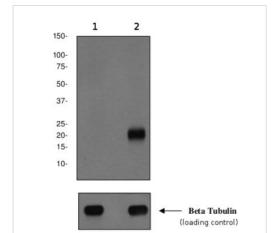
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 18 kDa
Observed band size: 18 kDa

Exposure time: 3 minutes

Blocking buffer 5% NFDM/TBST

Diluting buffer 5% NFDM/TBST



Western blot - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] (ab68236)

All lanes : Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] (ab68236) at 1/10000 dilution

Lane 1: Jurkat cell lysate, untreated.

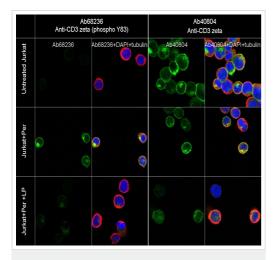
Lane 2: Jurkat cell lysate, treated with pervanadate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP-conjugated goat anti-rabbit lgG at 1/2000 dilution

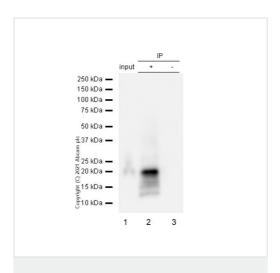
Predicted band size: 18 kDa **Observed band size:** 18-22 kDa



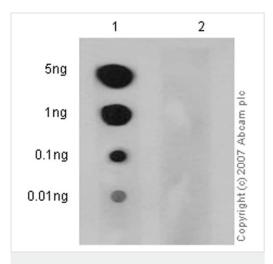
Immunocytochemistry/ Immunofluorescence - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] (ab68236) Immunocytochemistry/Immunofluorescence analysis of Jurkat cells (untreated, Per treated and Per+LP treated) labelling CD3 zeta (phospho Y83) with ab68236 (left) and CD3 zeta with ab40804 (right) both at a dilution of 1/200. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. ab7291, a mouse anti-tubulin (1/1000) and ab150120, an Alexa Fluor[®] 594-conjugated goat anti-mouse lgG (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.

<u>ab40804</u> 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.



Immunoprecipitation - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] (ab68236)



Dot Blot - Anti-CD3 zeta (phospho Y83) antibody [EP776(2)Y] (ab68236)

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 ab68236 at 1/30 dilution (2 μ g) . VeriBlot for IP Detection Reagent (HRP)(ab131366) 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

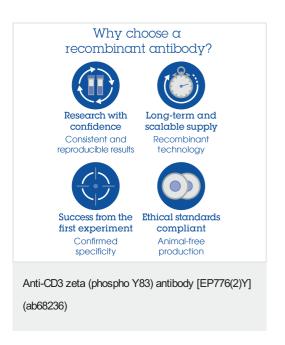
Lane 2: ab68236 IP in Jurkat treated with pervandate (50mM 5min) whole cell lysate

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab68236 in Jurkat treated with pervandate (50mM 5min) whole cell lysate Blocking and dilution buffer and concentration: 5% NFDM/TBST.

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.



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