abcam

Product datasheet

Anti-CD3 zeta antibody [BL-336-1B2] ab243874





7 Images

Overview

Product name Anti-CD3 zeta antibody [BL-336-1B2]

Description Rabbit monoclonal [BL-336-1B2] to CD3 zeta

Host species Rabbit

Tested applications Suitable for: WB, IHC-P, IP

Species reactivity Reacts with: Human

Immunogen Synthetic peptide within Human CD3 zeta aa 150-164 (C terminal). The exact sequence is

proprietary. NP_932170.1 and Gene ID 919.

Database link: P20963

Positive control WB: K562, Jurkat, HeLa, SR, HEK293T, HepG2 and MOLT4 whole cell lysate. IHC-P: Human

tonsil tissue, human lung cancer tissue. IP: Jurkat whole cell lysate.

General notes

This product is sold under License from Bethyl Laboratories, Inc.

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 long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.8

Preservative: 0.09% Sodium azide

Constituents: 98% Borate buffered saline, 0.1% BSA

Purification notes Recombinant antibody was purified from cell culture supernatant.

Clonality Monoclonal
Clone number BL-336-1B2

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab243874 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		1/1000.
IHC-P		1/100 - 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
IP		Use at an assay dependent concentration. Use 20µl/mg lysate.

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.

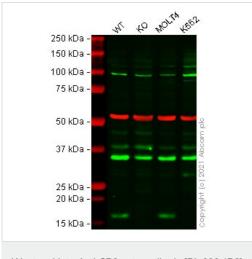
Domain The ITAM domains mediate interaction with SHB.

Post-translational Phosphorylated on Tyr residues after T-cell receptor triggering.

modifications

Cellular localization Membrane.

Images



Western blot - Anti-CD3 zeta antibody [BL-336-1B2] (ab243874)

All lanes: Anti-CD3 zeta antibody [BL-336-1B2] (ab243874) at

1/1000 dilution

Lane 1: Wild-type Jurkat cell lysate

Lane 2: CD247 knockout Jurkat cell lysate

Lane 3 : MOLT-4 cell lysate
Lane 4 : K562 cell lysate

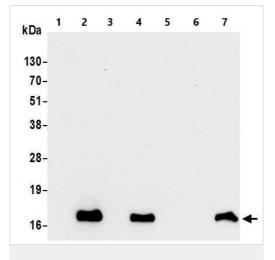
Lysates/proteins at 20 μg per lane.

Performed under reducing conditions.

Observed band size: 16 kDa

False colour image of Western blot: Anti-CD3 zeta antibody [BL-

336-1B2] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab243874 was shown to bind specifically to CD3 zeta. A band was observed at 16 kDa in wildtype Jurkat cell lysates with no signal observed at this size in CD247 knockout cell line ab273856 (knockout cell lysate ab273810). To generate this image, wild-type and CD247 knockout Jurkat cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3% milk in TBS-0.1% Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) at 1/20000 dilution.



Western blot - Anti-CD3 zeta antibody [BL-336-1B2] (ab243874)

All lanes : Anti-CD3 zeta antibody [BL-336-1B2] (ab243874) at 1/1000 dilution

Lane 1: K562 whole cell lysate

Lane 2: Jurkat whole cell lysate

Lane 3: HeLa whole cell lysate

Lane 4: SR whole cell lysate

Lane 5: HEK293T whole cell lysate

Lane 6: HepG2 whole cell lysate

Lane 7: MOLT4 whole cell lysate

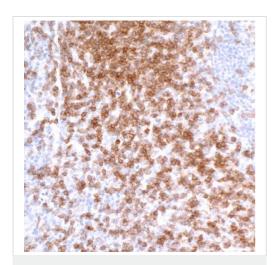
Lysates/proteins at 50 µg per lane.

Secondary

All lanes: HRP-conjugated goat anti-rabbit lgG

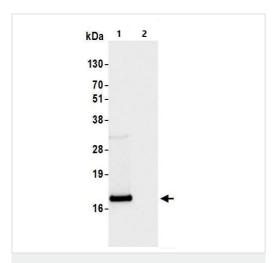
Exposure time: 10 seconds

Lysates prepared using NETN lysis buffer.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD3 zeta antibody [BL-336-1B2] (ab243874)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded human tonsil tissue labeling CD247/CD3Z with ab243874. HRP-conjugated goat anti-rabbit lgG was used as the secondary antibody, with DAB as substrate.



Immunoprecipitation - Anti-CD3 zeta antibody [BL-336-1B2] (ab243874)

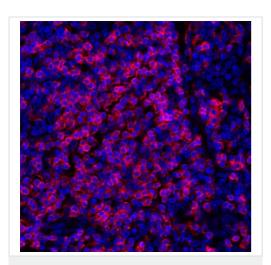
CD247/CD3Z was immunoprecipitated from 1mg of Jurkat whole cell lysate with ab243874 at 20 μ l/mg lysate. Western blot was performed from the immunoprecipitate using **ab264158** at a 1/1000 dilution.

Lane 1: ab243874 IP in Jurkat whole cell lysate.

Lane 2: Control IgG.

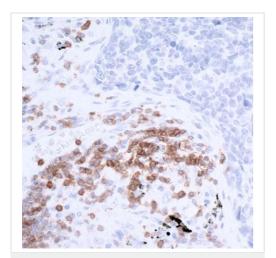
Detection: Chemiluminescence.

Lysate prepared using NETN lysis buffer.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD3 zeta antibody [BL-336-1B2] (ab243874)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded human tonsil tissue, labeling CD247/CD3Z with ab243874. HRP-conjugated goat anti-rabbit lgG used as the secondary antibody. DAPI counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD3 zeta antibody [BL-336-1B2] (ab243874)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded human lung cancer tissue labeling CD247/CD3Z with ab243874. HRP-conjugated goat anti-rabbit lgG was used as the secondary antibody, with DAB as the substrate.



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