abcam

Product datasheet

Anti-CD3D antibody [EPR20544] - BSA and Azide free ab229280



7 Images

Overview

Product name Anti-CD3D antibody [EPR20544] - BSA and Azide free

Description Rabbit monoclonal [EPR20544] to CD3D - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: WB, IHC-Fr, IP, Flow Cyt, IHC-P

Species reactivity Reacts with: Mouse

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

Positive control WB: Mouse thymus and lymph node lysates. IHC-P: Mouse spleen and thymus tissues. IHC-Fr:

Mouse spleen tissue. Flow Cyt: Mouse spleen cells. IP: Mouse thymus lysate.

General notes ab229280 is the carrier-free version of ab213362.

> Our carrier-free 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 cellbased 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**.

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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 numberEPR20544

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab229280 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		Use at an assay dependent concentration. Detects a band of approximately 24 kDa (predicted molecular weight: 19 kDa).
IHC-Fr		Use at an assay dependent concentration. Antigen retrieval: Heated citrate solution (10mM citrate pH 6.0 + 0.05% Tween-20).
IP		Use at an assay dependent concentration.
Flow Cyt		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Target

Function The CD3 complex mediates signal transduction.

Involvement in disease Defects in CD3D are a cause of severe combined immunodeficiency autosomal recessive T-cell-

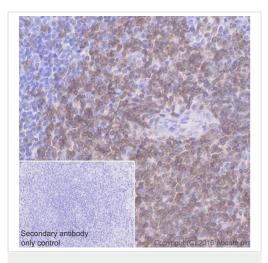
negative/B-cell-positive/NK-cell-positive (T(-)/B(+)/NK(+) SCID) [MIM:608971]. A form of severe combined immunodeficiency (SCID), a genetically and clinically heterogeneous group of rare congenital disorders characterized by impairment of both humoral and cell-mediated immunity, leukopenia, and low or absent antibody levels. Patients present in infancy recurrent, persistent infections by opportunistic organisms. The common characteristic of all types of SCID is absence

of T-cell-mediated cellular immunity due to a defect in T-cell development.

Sequence similarities Contains 1 ITAM domain.

Cellular localization Membrane.

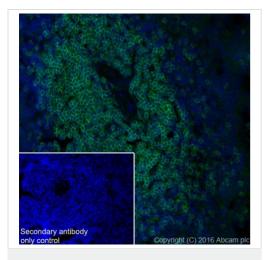
Images



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD3D antibody
[EPR20544] - BSA and Azide free (ab229280)

This data was developed using <u>ab213362</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling CD3D with <u>ab213362</u> at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic staining on T cells of mouse splenic periarterial lymphatic sheath is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Frozen sections) - Anti-CD3D antibody [EPR20544] - BSA and Azide free (ab229280)

This data was developed using <u>ab213362</u>, the same antibody clone in a different buffer formulation.lmmunohistochemical analysis of 4% paraformaldehyde-fixed, 0.2% Triton X-100 permeabilized frozen mouse spleen tissue labeling CD3D with <u>ab213362</u> at 1/100 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor[®] 488) (<u>ab150077</u>) secondary antibody at 1/1000 dilution (green). Cytoplasmic and membranous staining on mouse splenic periarterial lymphatic sheath is observed. The nuclear counterstain is DAPI (blue). Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit IgG (Alexa Fluor[®] 488) (<u>ab150077</u>) at 1/1000 dilution.



Western blot - Anti-CD3D antibody [EPR20544] - BSA and Azide free (ab229280)

All lanes : Anti-CD3D antibody [EPR20544] (<u>ab213362</u>) at 1/1000 dilution

Lane 1: Mouse thymus tissue lysate

Lane 2: Mouse lymph node tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 19 kDa **Observed band size:** 24 kDa

This data was developed using <u>ab213362</u>, the same antibody clone in a different buffer formulation.

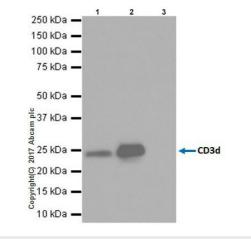
Blocking and dilution buffer: 5% NFDM/TBST.

Exposure times: Lane 1: 15 seconds; Lane 2: 3 minutes.

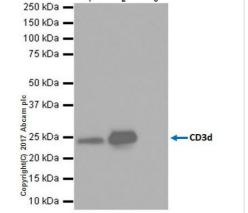
| Sotype Control - Alexa Fluor® 488 (530/30 BP) | CD3d - Alexa Fluor® 488 (530/30 BP)

Flow Cytometry - Anti-CD3D antibody [EPR20544] - BSA and Azide free (ab229280)

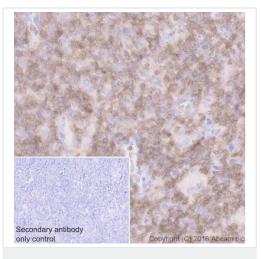
This data was developed using <u>ab213362</u>, the same antibody clone in a different buffer formulation. Flow cytometric analysis of mouse spleen cells labeling CD3D with <u>ab213362</u> at 1/600 dilution (right panel), compared with a rabbit monoclonal IgG isotype control (<u>ab172730</u>) (left panel). Goat anti rabbit IgG (Alexa Fluor[®] 488) at 1/2000 dilution was used as the secondary antibody. Firstly, cells surface was stained with anti-mouse CD3 Alexa Fluor 647 (Y axis), then cells were fixed with 4% PFA followed by intracellular staining with <u>ab213362</u>. The same population of cells were stained by both: anti-mouse CD3 and anti-mouse CD3 (<u>ab213362</u>) antibodies.



Immunoprecipitation - Anti-CD3D antibody



[EPR20544] - BSA and Azide free (ab229280)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD3D antibody [EPR20544] - BSA and Azide free (ab229280)

This data was developed using ab213362, the same antibody clone in a different buffer formulation.

CD3D was immunoprecipitated from 0.35 mg of mouse thymus lysate with ab213362 at 1/30 dilution.

Western blot was performed from the immunoprecipitate using ab213362 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10,000 dilution.

Lane 1: Mouse thymus lysate, 10 µg (Input).

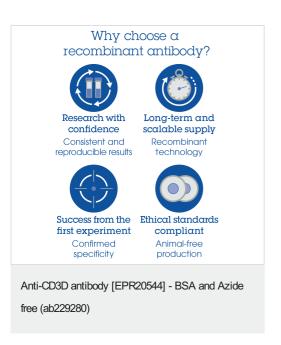
Lane 2: ab213362 IP in mouse thymus lysate.

Lane 3: Rabbit monoclonal lgG (ab172730) instead of ab213362 in mouse thymus lysate.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 10 seconds.

This data was developed using ab213362, the same antibody clone in a different buffer formulation.lmmunohistochemical analysis of paraffin-embedded mouse thymus tissue labeling CD3D with ab213362 at 1/2000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) Ready to use. Cytoplasmic and membranous staining on T cells of mouse thymus medulla is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



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