abcam

Product datasheet

Anti-CD3D antibody [EPR20544] ab213362



7 Images

Overview

Product name Anti-CD3D antibody [EPR20544]

Description Rabbit monoclonal [EPR20544] to CD3D

Host species Rabbit

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

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 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**.

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: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number EPR20544

Isotype ΙgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab213362 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. Detects a band of approximately 24 kDa (predicted molecular weight: 19 kDa).
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IHC-Fr		1/100. Antigen retrieval: Heated citrate solution (10mM citrate pH 6.0 + 0.05% Tween-20).
Flow Cyt		1/600.
IP		1/30.

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

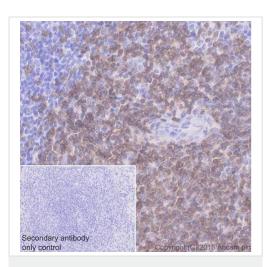
Contains 1 ITAM domain.

Cellular localization

Membrane.

Form

Images



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD3D antibody
[EPR20544] (ab213362)

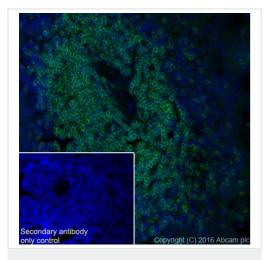
Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling CD3D with ab213362 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

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 lgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



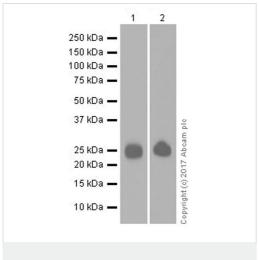
Immunohistochemistry (Frozen sections) - Anti-CD3D antibody [EPR20544] (ab213362)

Immunohistochemical analysis of 4% paraformaldehyde-fixed, 0.2% Triton X-100 permeabilized frozen mouse spleen tissue labeling CD3D with ab213362 at 1/100 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor[®] 488) (**ab150077**) 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 lgG (Alexa Fluor[®] 488) (ab150077) at 1/1000 dilution.



Western blot - Anti-CD3D antibody [EPR20544] (ab213362)

All lanes : Anti-CD3D antibody [EPR20544] (ab213362) 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) (ab97051) at

1/100000 dilution

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

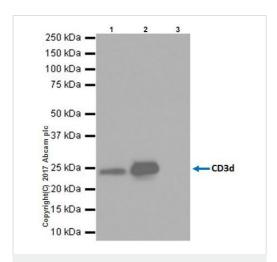
Blocking/Dilution buffer: 5% NFDM/TBST.

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

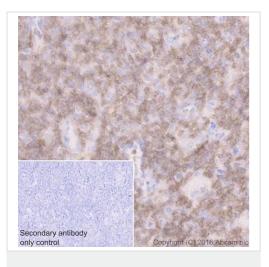
Flow Cytometry - Anti-CD3D antibody [EPR20544] (ab213362)

Flow cytometric analysis of mouse spleen cells labeling CD3D with ab213362 at 1/600 dilution (right panel), compared with a rabbit monoclonal lgG isotype control (<u>ab172730</u>) (left panel). Goat anti rabbit lgG (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 ab213362. The same population of cells were stained by both: anti-mouse CD3 and anti-mouse CD3D (ab213362) antibodies.



Immunoprecipitation - Anti-CD3D antibody [EPR20544] (ab213362)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD3D antibody
[EPR20544] (ab213362)

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) (<u>ab131366</u>), 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 $\lg G$ ($\underline{ab172730}$) instead of ab213362 in mouse thymus lysate .

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

Exposure time: 10 seconds.

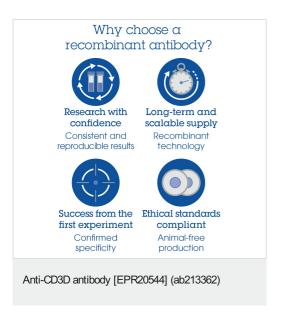
Immunohistochemical analysis of paraffin-embedded mouse thymus tissue labeling CD3D with ab213362 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

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 lgG 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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