abcam

Product datasheet

Anti-FADD antibody [EPR5030] ab124812



★★★★ 9 Abreviews 23 References

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Overview

Product name Anti-FADD antibody [EPR5030]

Description Rabbit monoclonal [EPR5030] to FADD

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IHC-P, IP, ICC/IF

Species reactivity Reacts with: Mouse

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

Positive control IHC-P: Mouse kidney tissue; ICC/IF: NIH/3T3 cells; Flow Cyt (intra): NIH/3T3 cells. WB: NIH/3T3

whole cell lysate. IP: NIH/3T3 whole cell lysate and RAW264.7 whole cell 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**.

Human, 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 short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

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

Purity Protein A purified

Clonality Monoclonal

Clone number **EPR5030**

Isotype ΙgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab124812 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)		1/40. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB	★★★★	1/1000 - 1/10000. Detects a band of approximately 28 kDa (predicted molecular weight: 23 kDa).
IHC-P		1/50 - 1/150. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols.
IP	★★★★ <u>(1)</u>	1/30.
ICC/IF		1/50.

Target

Function Apoptotic adaptor molecule that recruits caspase-8 or caspase-10 to the activated Fas (CD95)

> or TNFR-1 receptors. The resulting aggregate called the death-inducing signaling complex (DISC) performs caspase-8 proteolytic activation. Active caspase-8 initiates the subsequent cascade of

caspases mediating apoptosis.

Tissue specificity Expressed in a wide variety of tissues, except for peripheral blood mononuclear leukocytes.

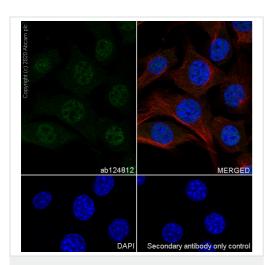
Sequence similarities Contains 1 death domain.

Contains 1 DED (death effector) domain.

Domain Contains a death domain involved in the binding of the corresponding domain within Fas

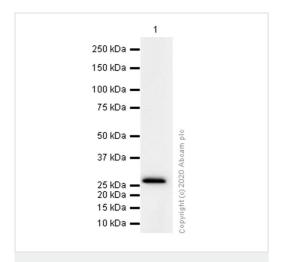
receptor.

Images



Immunocytochemistry/ Immunofluorescence - Anti-FADD antibody [EPR5030] (ab124812)

Immunocytochemistry analysis of NIH/3T3 (Mouse embryonic fibroblast) cells labeling FADD with purified ab124812 at 1/50 dilution (7.8 µg/mL). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor $^{@}$ 594) 1/200 (2.5 µg/mL). Goat anti rabbit lgG (Alexa Fluor $^{@}$ 488, <code>ab150077</code>) was used as the secondary antibody at 1/1000 (2 µg/mL) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



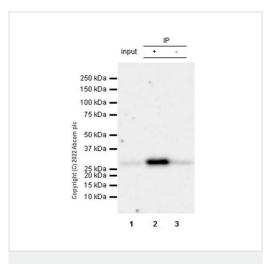
Western blot - Anti-FADD antibody [EPR5030] (ab124812)

Anti-FADD antibody [EPR5030] (ab124812) at 1/1000 dilution (Purified) + NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 23 kDa



Immunoprecipitation - Anti-FADD antibody [EPR5030] (ab124812)

FADD was immunoprecipitated from 0.35 mg NIH/3T3 (mouse embryonic fibroblast) whole cell lysate 10 μ g with ab124812 at 1/30 dilution (2 μ g in 0.35 mg lysates). Western blot was performed on the immunoprecipitate using ab124812 at 1/1000 dilution. VeriBlot for IP secondary antibody (HRP) (ab131366) was used at 1/5000 dilution.

Lane 1: NIH/3T3 (mouse embryonic fibroblast) whole cell lysate 10 µg.

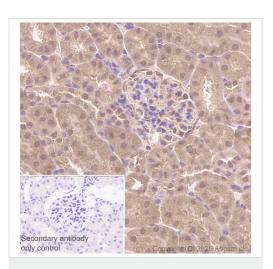
Lane 2: NIH/3T3 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab124812 in NIH/3T3 whole cell lysate.

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

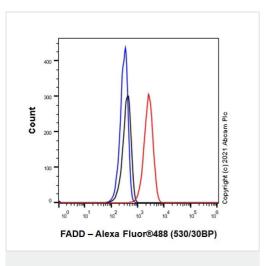
Observed MW: 28 kDa.

Exposure time: 41 secs.



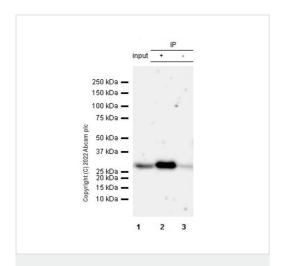
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-FADD antibody
[EPR5030] (ab124812)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse kidney tissue sections labeling FADD with purified ab124812 at 1/150 dilution (2.59 µg/mL). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1/0 dilution. PBS instead of the primary antibody was used as the negative control.



Flow Cytometry (Intracellular) - Anti-FADD antibody [EPR5030] (ab124812)

Intracellular Flow Cytometry analysis of NIH/3T3 (Mouse embryonic fibroblast) cells labelling FADD with purified ab124812 at 1/40 dilution (10 μ g/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor 488, ab150077) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunoprecipitation - Anti-FADD antibody [EPR5030] (ab124812)

FADD was immunoprecipitated from 0.35 mg RAW264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate 10 μ g with ab124812 at 1/30 dilution (2 μ g in 0.35 mg lysates). Western blot was performed on the immunoprecipitate using ab124812 at 1/1000 dilution. VeriBlot for IP secondary antibody (HRP) (ab131366) was used at 1/5000 dilution.

Lane 1: RAW264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate 10 µg.

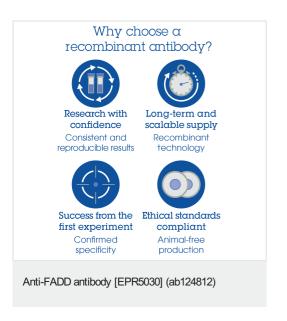
Lane 2: RAW264.7 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab124812 in RAW264.7 whole cell lysate.

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

Observed MW: 28 kDa.

Exposure time: 3 minutes.



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