abcam

Product datasheet

Anti-Ibal antibody [EPR16589] - BSA and Azide free ab221790





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Overview

Product name Anti-lba1 antibody [EPR16589] - BSA and Azide free

Description Rabbit monoclonal [EPR16589] to lba1 - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: IHC-P, ICC/IF, IP, WB, IHC (PFA fixed)

Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control WB: Human spleen lysate; THP-1, MOLT-4 and U937 whole cell lysates; Mouse and rat spleen

and testis lysates. IHC-P: Human cerebrum, mouse endometrium and rat cerebrum tissues.

ICC/IF: U937 and THP-1 cells. IP: Mouse spleen whole cell lysate.

General notes ab221790 is the carrier-free version of ab178847.

> 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**.

1

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

Clonality Monoclonal
Clone number EPR16589

Isotype IgG

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab221790 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
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.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 17 kDa (predicted molecular weight: 17 kDa).
IHC (PFA fixed)		Use at an assay dependent concentration.

Target

Function Actin-binding protein that enhances membrane ruffling and RAC activation. Enhances the actin-

bundling activity of LCP1. Binds calcium. Plays a role in RAC signaling and in phagocytosis. May play a role in macrophage activation and function. Promotes the proliferation of vascular smooth muscle cells and of T-lymphocytes. Enhances lymphocyte migration. Plays a role in vascular

inflammation.

Tissue specificity Detected in T-lymphocytes and peripheral blood mononuclear cells.

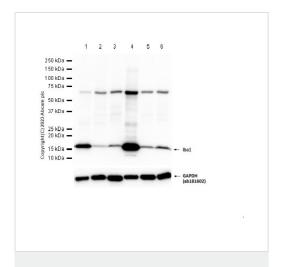
Sequence similarities Contains 2 EF-hand domains.

Post-translational Phosphorylated on serine residues.

modifications

Cellular localization Cytoplasm > cytoskeleton. Cell projection > ruffle membrane. Associated with the actin

Images



Western blot - Anti-lba1 antibody [EPR16589] - BSA and Azide free (ab221790)

All lanes : Anti-lba1 antibody [EPR16589] (<u>ab178847</u>) at 1/1000 dilution

Lane 1: Mouse spleen tissue lysate

Lane 2: Mouse brain tissue lysate

Lane 3: Mouse hippocampus tissue lysate

Lane 4: Rat spleen tissue lysate

Lane 5: Rat brain tissue lysate

Lane 6: Rat hippocampus tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

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

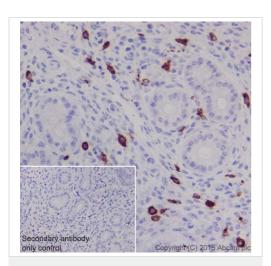
Predicted band size: 17 kDa **Observed band size:** 17 kDa

Exposure time: 40 seconds

This data was developed using <u>ab178847</u>, the same antibody clone in a different buffer formulation.

Blocking/Dilution buffer: 5% NFDM/TBST.

IBA1 is a relatively minor protein of brain and is much more abundant in spleen (PMID: 8912632, PMID: 29232670). We suggest loading higher amount of brain lysate or using lower dilution of antibody for detecting signal in brain related lysates.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Iba1 antibody

[EPR16589] - BSA and Azide free (ab221790)

ab178847 MERGED

OPPIN

Immunocytochemistry/ Immunofluorescence - Anti-Iba1 antibody [EPR16589] - BSA and Azide free (ab221790)

Immunohistochemical analysis of paraffin-embedded Mouse endometrium tissue labeling lba1 with <u>ab178847</u> at 1/8000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

Cytoplasm staining on macrophages of the mouse endometrium.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab178847).

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

Immunofluorescent analysis of 100% methanol-fixed, 0.1% Triton X-100 permeabilized U937 (Human histiocytic lymphoma cell line) cells labeling lba1 with ab178847 at 1/100 dilution, followed by Goat Anti-Rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing cytoplasmic staining on U937 cell line.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin - Loading
Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse IgG
(AlexaFluor®594) preadsorbed (ab150120) at 1/1000 dilution (red).

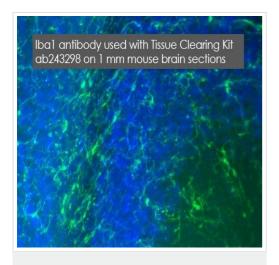
The negative controls are as follows:-

-ve control 1: <u>ab178847</u> at 1/100 dilution followed by <u>ab150120</u> at 1/1000 dilution.

-ve control 2: <u>ab7291</u> at 1/1000 dilution followed by <u>ab150077</u> at 1/1000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and

sodium azide (ab178847).



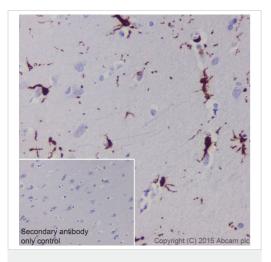
Immunohistochemistry (PFA fixed) - Anti-lba1 antibody [EPR16589] - BSA and Azide free (ab221790)

This data was developed using <u>ab178847</u>, the same antibody clone in a different buffer formulation.

lba1 antibody <u>ab178847</u> was used with Tissue Clearing
Kit <u>ab243298</u> to penetrate, stain and clear a 1 mm coronal section
of mouse brain. Blue: DAPI, Green: lba1.

Learn more about <u>tissue clearing kits, reagents, and</u>
<u>protocols</u> designed to make it easier to stain thick tissue sections and get more data from each valuable tissue section.

For 1 mm brain sections, we recommend a starting dilution of 1:100, and also using Goat Anti-Rabbit lgG H&L AlexaFluor488 (ab150077) at a dilution of 1:400.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Iba1 antibody

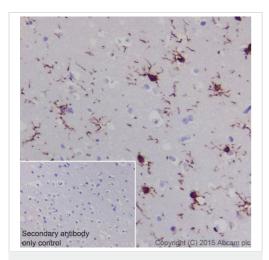
[EPR16589] - BSA and Azide free (ab221790)

Immunohistochemical analysis of paraffin-embedded
Human cerebrum tissue labeling lba1 with <u>ab178847</u> at 1/8000
dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

Cytoplasm staining on microglia of the normal Human cerebrum is observed. Counter stained with Hematoxylin.

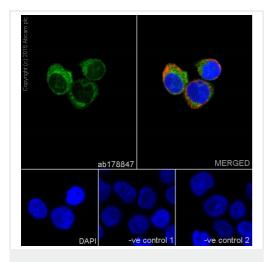
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is <u>ab97051</u> at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab178847).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Iba1 antibody

[EPR16589] - BSA and Azide free (ab221790)



Immunocytochemistry/ Immunofluorescence - Anti-Iba1 antibody [EPR16589] - BSA and Azide free (ab221790)

Immunohistochemical analysis of paraffin-embedded Rat cerebrum tissue labeling lba1 with <u>ab178847</u> at 1/8000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

Cytoplasm staining on microglia of the rat cerebrum 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) (ab97051) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab178847).

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

Immunofluorescent analysis of 100% methanol-fixed, 0.1% Triton X-100 permeabilized THP-1 (Human monocytic leukemia cell line) cells labeling lba1 with ab178847 at 1/100 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing cytoplasmic staining on THP-1 cell line.

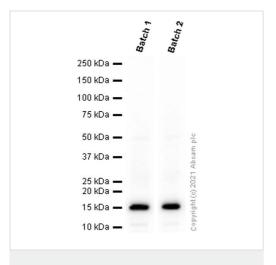
The nuclear counterstain is DAPI (blue). Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution and <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution (red).

The negative controls are as follows:-

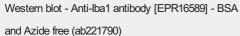
-ve control 1: $\underline{ab178847}$ at 1/100 dilution followed by $\underline{ab150120}$ at 1/1000 dilution.

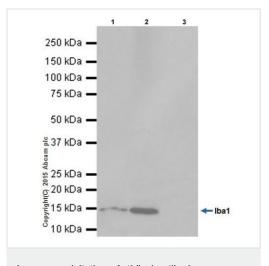
-ve control 2: $\underline{ab7291}$ at 1/1000 dilution followed by $\underline{ab150077}$ at 1/1000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab178847</u>).



This data was developed using <u>ab178847</u>, the same antibody clone in a different buffer formulation. Different batches of <u>ab178847</u> were tested on U-937 (Human histiocytic lymphoma monocyte) lysate at $0.5~\mu g/ml$. $15~\mu g$ of lysate was loaded in each lane. Bands observed at 17~kDa.





Immunoprecipitation - Anti-Iba1 antibody [EPR16589] - BSA and Azide free (ab221790) lba1 was immunoprecipitated from 1mg of Mouse spleen whole cell lysate with ab178847 at 1/40 dilution.

Western blot was performed from the immunoprecipitate using <u>ab178847</u> at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>), was used for detection at 1/10000 dilution.

Lane 1: Mouse spleen whole cell lysate 10µg (Input).

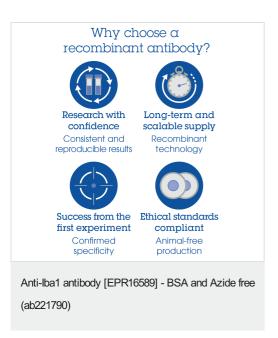
Lane 2: $\underline{ab178847}$ IP in Mouse spleen whole cell lysate.

Lane 3: Rabbit lgG,monoclonal-Isotype Control (<u>ab172730</u>) instead of <u>ab178847</u> in Mouse spleen whole cell lysate.

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

Exposure time: 5 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab178847).



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