abcam

Product datasheet

Anti-Profilin 1 antibody [EPR6304] ab124904





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Overview

Product name Anti-Profilin 1 antibody [EPR6304]

Description Rabbit monoclonal [EPR6304] to Profilin 1

Host species Rabbit

Specificity The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for

mouse and rat.

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

Species reactivity Reacts with: Mouse, Rat, Human

Synthetic peptide within Human Profilin 1 aa 100-200 (C terminal). The exact sequence is **Immunogen**

proprietary.

Positive control WB: Jurkat, JAR, HepG2 and HEK-293T cell lysates, mouse and rat brain lysate, human fetal

brain lysate; IHC-P: Human colon and breast tissue; ICC/IF: HeLa, Jurkat cells; Flow Cyt (intra):

HeLa cells:

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**.

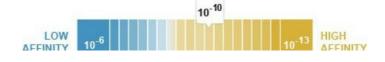
Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Stable for 12 months at -20°C.

 $K_D = 1.24 \times 10^{-10} M$ Dissociation constant (K_D)





Learn more about K_D

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number EPR6304

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab124904 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)	1/10000 - 1/50000. Detects a band of approximately 12 kDa (predicted molecular weight: 15 kDa).
IHC-P		1/500 - 1/1000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat. See IHC antigen retrieval protocols.
ICC/IF		1/100. For unpurified use at 1/250 - 1/500
Flow Cyt (Intra)		1/200. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

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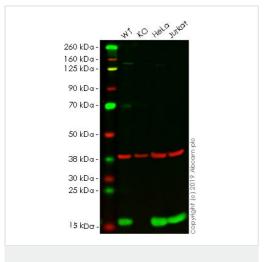
Function Binds to actin and affects the structure of the cytoskeleton. At high concentrations, profilin

prevents the polymerization of actin, whereas it enhances it at low concentrations. By binding to

PIP2, it inhibits the formation of IP3 and DG.

Sequence similaritiesBelongs to the profilin family.Cellular localizationCytoplasm > cytoskeleton.

Images



Western blot - Anti-Profilin 1 antibody [EPR6304] (ab124904)

All lanes : Anti-Profilin 1 antibody [EPR6304] (ab124904) at 1/1000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: PFN1 knockout HAP1 whole cell lysate

Lane 3 : HeLa whole cell lysate

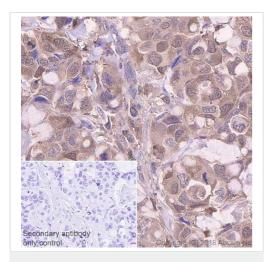
Lane 4 : Jurkat whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 15 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab124904 observed at 15 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

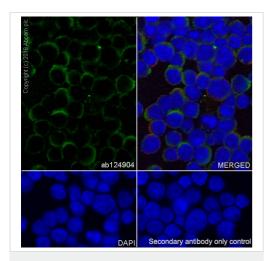
ab124904 was shown to specifically react with in wild-type HAP1 cells as signal was lost in PFN1 knockout cells. Wild-type and PFN1 knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% NF Milk. Ab124904 and <u>ab8245</u> (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/10000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed <u>ab216776</u> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Profilin 1 antibody

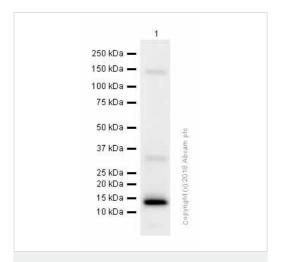
[EPR6304] (ab124904)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast carcinoma tissue sections labeling Profilin 1 with Purified ab124904 at 1:800 dilution (0.18 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunocytochemistry/ Immunofluorescence - Anti-Profilin 1 antibody [EPR6304] (ab124904)

Immunocytochemistry/ Immunofluorescence analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling Profilin 1 with Purified ab124904 at 1:100 dilution (1.4 μ g/ml). Cells were fixed in 100% Methanol. Cells were counterstained with Ab195889 Antialpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 μ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1:1000 (2 μ g/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



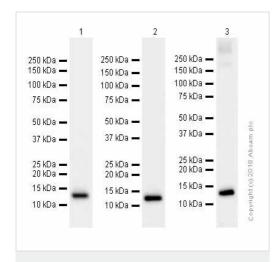
Western blot - Anti-Profilin 1 antibody [EPR6304] (ab124904)

Anti-Profilin 1 antibody [EPR6304] (ab124904) at 1/10000 dilution (Purified) + Human fetal brain lysates at 15 µg

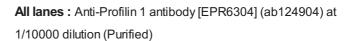
Secondary

Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 15 kDa **Observed band size:** 12 kDa



Western blot - Anti-Profilin 1 antibody [EPR6304] (ab124904)



Lane 1 : Jurkat (Human T cell leukemia T lymphocyte) whole cell lysates

Lane 2: Mouse brain lysates

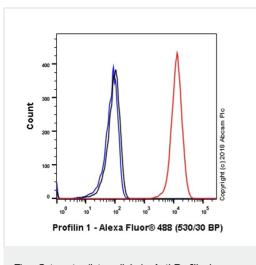
Lane 3: Rat brain lysates

Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/20000 dilution

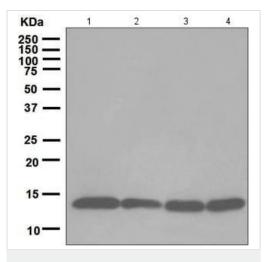
Predicted band size: 15 kDa **Observed band size:** 12 kDa



Flow Cytometry (Intracellular) - Anti-Profilin 1 antibody [EPR6304] (ab124904)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Profilin 1 with Purified ab124904 at 1/200 dilution (1 μ g/ml) (red). Cells were fixed with 80% Methanol. A Goat anti rabbit lgG (Alexa Fluor $^{\otimes}$ 488,

<u>ab150077</u>) secondary antibody was used at 1/2000. Isotype control
 Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Western blot - Anti-Profilin 1 antibody [EPR6304] (ab124904)

All lanes : Anti-Profilin 1 antibody [EPR6304] (ab124904) at 1/10000 dilution (Unpurified)

Lane 1 : Jurkat cell lysate

Lane 2 : JAR cell lysate

Lane 3: HepG2 cell lysate

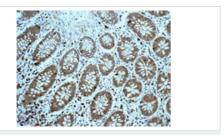
Lane 4: 293T cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP labelled goat anti-rabbit at 1/2000 dilution

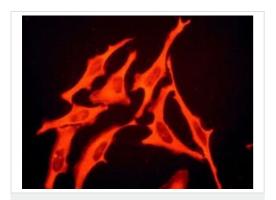
Predicted band size: 15 kDa **Observed band size:** 12 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Profilin 1 antibody
[EPR6304] (ab124904)

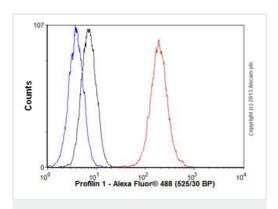
Unpurified ab124904, at 1/500 dilution, staining Profilin 1 in paraffin-embedded human colon tissue by Immunohistochemistry.

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



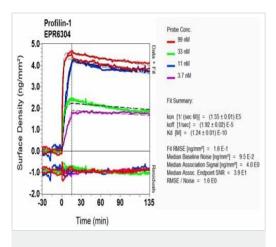
Immunocytochemistry/ Immunofluorescence - Anti-Profilin 1 antibody [EPR6304] (ab124904)

Unpurified ab124904, at 1/250 dilution, staining Profilin 1 in HeLa cells by Immunofluorescence.



Flow Cytometry (Intracellular) - Anti-Profilin 1 antibody [EPR6304] (ab124904)

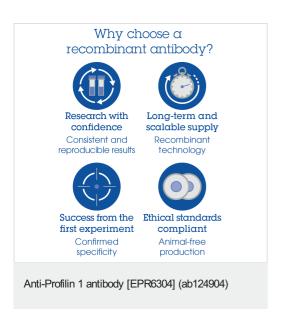
Overlay histogram showing HeLa cells stained with unpurifiedab124904 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab124904, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor 488 goat anti-rabbit lgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (0.1 μ g/1x10 6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



OI-RD Scanning - Anti-Profilin 1 antibody [EPR6304] (ab124904)

Equilibrium disassociation constant (K_D) Learn more about K_D

Click here to learn more about $K_{\underline{D}}$



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