abcam

Product datasheet

Anti-IL-18 antibody [EPR19954] ab207324



Recombinant RabMAb

8 References 9 Images

Overview

Product name Anti-IL-18 antibody [EPR19954]

Rabbit monoclonal [EPR19954] to IL-18 **Description**

Host species Rabbit

Tested applications Suitable for: WB, Flow Cyt (Intra)

Species reactivity Reacts with: Human

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Human IL-18 active protein; HeLa, A431, Jurkat, HEK-293, PC-3 and HaCaT whole cell

lysates; Human skin and ovary cancer lysates. Flow Cyt (intra): PC-3 and HaCaT 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 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 EPR19954

Isotype ΙgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab207324 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		1/200. Detects a band of approximately 22 kDa (predicted molecular weight: 22 kDa).
Flow Cyt (Intra)		1/60.

Target

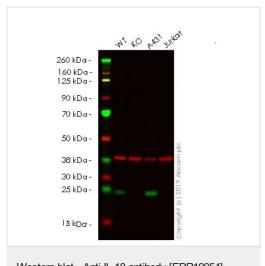
Function Augments natural killer cell activity in spleen cells and stimulates interferon gamma production in

T-helper type I cells.

Sequence similarities Belongs to the IL-1 family.

Cellular localization Secreted.

Images



Western blot - Anti-IL-18 antibody [EPR19954] (ab207324)

All lanes : Anti-IL-18 antibody [EPR19954] (ab207324) at 1/200

dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: IL18 knockout HeLa cell lysate

Lane 3 : A431 cell lysate

Lane 4 : Jurkat cell lysate

Lysates/proteins at 20 µg per lane.

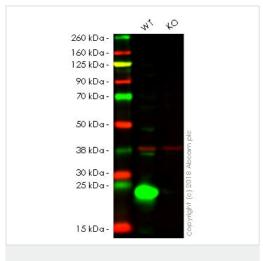
Performed under reducing conditions.

Predicted band size: 22 kDa Observed band size: 22 kDa

Lanes 1-4: Merged signal (red and green). Green - ab207324 observed at 22 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab207324 Anti-IL-18 antibody [EPR19954] was shown to specifically react with IL-18 in wild-type HeLa cells. Loss of signal

was observed when knockout cell line ab256952) was used. Wild-type and IL-18 knockout samples were subjected to SDS-PAGE. ab207324 and Anti-alpha Tubulin antibody [EP1332Y] - Microtubule Marker (ab52866) were incubated overnight at 4°C at 1 in 200 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-IL-18 antibody [EPR19954] (ab207324)

All lanes : Anti-IL-18 antibody [EPR19954] (ab207324) at 1/200 dilution

Lane 1: Wild-type HEK-293 whole cell lysate

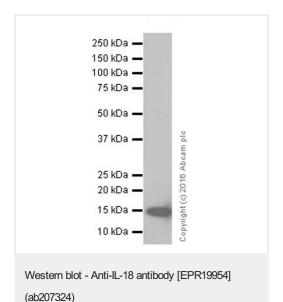
Lane 2: IL-18 knockout HEK-293 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 22 kDa

Lanes 1 - 2: Merged signal (red and green). Green - ab207324 observed at 22 kDa. Red - loading control, <u>ab9484</u>, observed at 37 kDa.

ab207324 was shown to recognize IL-18 in wild-type HEK293 cells as signal was lost at the expected MW in IL-18 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and IL-18 knockout samples were subjected to SDS-PAGE. Ab207324 and ab9484 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/200 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Anti-IL-18 antibody [EPR19954] (ab207324) at 1/1000 dilution + Human IL-18 active protein at 0.02 µg

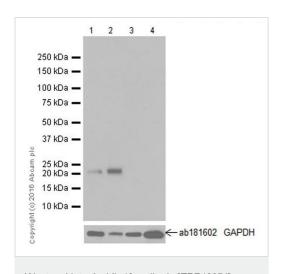
Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 22 kDa **Observed band size:** 17 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-IL-18 antibody [EPR19954] (ab207324)

All lanes : Anti-IL-18 antibody [EPR19954] (ab207324) at 1/1000 dilution

Lane 1 : PC-3 (Human prostate adenocarcinoma cell line) whole cell lysate

Lane 2: HaCaT (Human keratinocyte cell line) whole cell lysate

Lane 3: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 4 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/50000 dilution

Predicted band size: 22 kDa **Observed band size:** 22 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

2 250 kDa -250 kDa -150 kDa -150 kDa -100 kDa -100 kDa -75 kDa -75 kDa -50 kDa -Copyright (c) 2016 Abcam plo 50 kDa -37 kDa -37 kDa -25 kDa -25 kDa — 20 kDa — 20 kDa -15 kDa -15 kDa -10 kDa -10 kDa -

Western blot - Anti-IL-18 antibody [EPR19954] (ab207324)

The WB profiles are consistent with the literature (PMID 12918059; PMID 11470273).

All lanes : Anti-IL-18 antibody [EPR19954] (ab207324) at 1/200 dilution

Lane 1: Human skin lysate

Lane 2: Human ovary cancer lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/5000 dilution

Predicted band size: 22 kDa Observed band size: 22 kDa

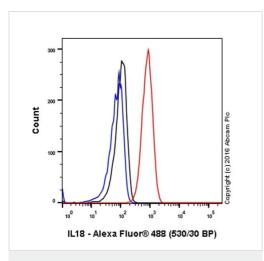
Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

Detection in skin and ovary required high concentration of antibody.

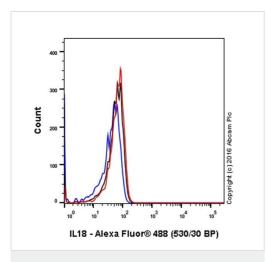
IL18- Alexa Fluor® 488 (530/30 BP)

Flow Cytometry (Intracellular) - Anti-IL-18 antibody [EPR19954] (ab207324) Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed PC-3 (Human prostate adenocarcinoma cell line) cells labeling IL-18with ab207324 at 1/60 dilution (red) compared with Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat Anti-Rabbit IgG (Alexa Fluorr[®] 488) at 1/2000 dilution was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-IL-18 antibody [EPR19954] (ab207324)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HaCaT (Human keratinocyte cell line) cells labeling IL-18with ab207324 at 1/60 dilution (red) compared with Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab172730; black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody; blue). Goat Anti-Rabbit IgG (Alexa Fluorr® 488) at 1/2000 dilution was used as the secondary antibody.

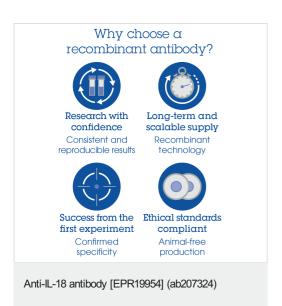


Flow Cytometry (Intracellular) - Anti-IL-18 antibody [EPR19954] (ab207324)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed Jurkat (Human T cell leukemia cell line from peripheral blood) cells labeling IL-18with ab207324 at 1/60 dilution (red) compared with a rabbit monoclonal IgG isotype control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (Alexa Fluorr® 488) at 1/2000 dilution was used as the secondary antibody.

Negative control:

Jurkat cells serve as a negative cell line as described in PMID 15086390.



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