abcam

Product datasheet

Anti-CXCL9 antibody [EPR23999-5] ab263442





6 Images

Overview

Product name Anti-CXCL9 antibody [EPR23999-5]

Description Rabbit monoclonal [EPR23999-5] to CXCL9

Host species Rabbit

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

Unsuitable for: ICC/IF or IHC-P

Reacts with: Human Species reactivity

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

Positive control WB: THP-1 (treated with 200ng/ml IFN-? and 50 ng/ml LPS(Lipopolysaccharide) for 24 hours)

> whole cell lysate; His-tagged human purified recombinant full-length protein CXCL9. Flow Cyt (intra): THP-1 (treated with 100ng/ml IFN-gamma and 1ug/ml LPS(Lipopolysaccharide) for 48 hours) cells. IP: THP-1 (treated with 100ng/ml IFN-gamma and 1ug/ml LPS(Lipopolysaccharide)

for 48 hours) 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**.

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 Preservative: 0.01% Sodium azide

Constituents: 59.94% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal

Clone number

EPR23999-5

Isotype

lgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab263442 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)		1/500.
WB		1/1000. Detects a band of approximately 13, 14 kDa (predicted molecular weight: 14 kDa).
IP		1/30.

Application notes

Is unsuitable for ICC/IF or IHC-P.

Target

Function

Cytokine that affects the growth, movement, or activation state of cells that participate in immune and inflammatory response. Chemotactic for activated T-cells. Binds to CXCR3.

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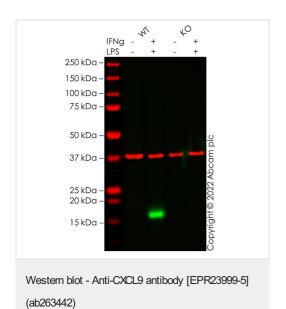
Sequence similarities

Belongs to the intercrine alpha (chemokine CxC) family.

Cellular localization

Secreted.

Images



All lanes : Anti-CXCL9 antibody [EPR23999-5] (ab263442) at 1/1000 dilution

Lane 1: Wild-type THP-1 untreated control cell lysate

Lane 2: Wild-type THP-1 treated IFNg (200 ng/mL, 24 h) + LPS

(50 ng/mL, 24 h) cell lysate

Lane 3: CXCL9 knockout THP-1 untreated control cell lysate

Lane 4: CXCL9 knockout THP-1 treated IFNg (200 ng/mL, 24 h) +

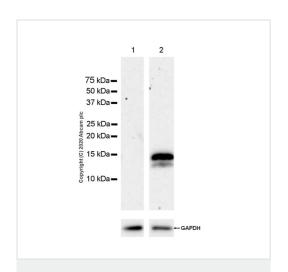
LPS (50 ng/mL, 24 h) cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 14 kDa **Observed band size:** 17 kDa

False colour image of Western blot: Anti-CXCL9 antibody [EPR23999-5] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab263442 was shown to bind specifically to CXCL9. A band was observed at 17 kDa in treated wild-type THP-1 cell lysates with no signal observed at this size in CXCL9 knockout cell line ab282347 (knockout cell lysate ab283019). To generate this image, wild-type and CXCL9 knockout THP-1 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.



Western blot - Anti-CXCL9 antibody [EPR23999-5] (ab263442)

All lanes : Anti-CXCL9 antibody [EPR23999-5] (ab263442) at 1/1000 dilution

Lane 1 : Untreated THP-1 (human monocytic leukemia monocyte), whole cell lysate

Lane 2: THP-1 (treated with 200 ng /ml IFN-gamma and 50 ng/ml LPS (Lipopolysaccharide) for 24 hours), whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

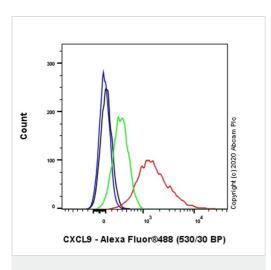
All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 14 kDa **Observed band size:** 13,14 kDa

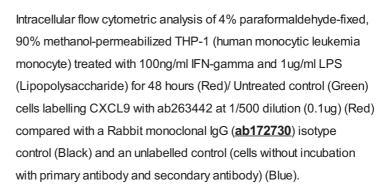
Blocking and diluting buffer and concentration: 5% NFDM/TBST.

The expression profile is consistent with what has been described in the literature (PMID: 15154616, 22973274).

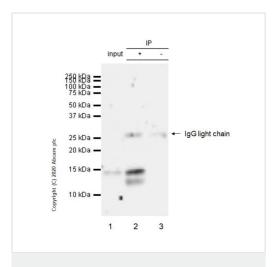
Exposure time: 3 minutes.



Flow Cytometry (Intracellular) - Anti-CXCL9 antibody [EPR23999-5] (ab263442)



A Goat anti rabbit lgG (Alexa Fluor[®] 488, <u>ab150077</u>) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-CXCL9 antibody [EPR23999-5] (ab263442)

CXCL9 was immunoprecipitated from 0.35 mg THP-1(human monocytic leukemia monocyte) (treated with 100ng/ml IFN-γ and 1ug/ml LPS (Lipopolysaccharide) for 48 hours) whole cell lysate with ab263442 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab263442 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366) was used at 1/5000 dilution.

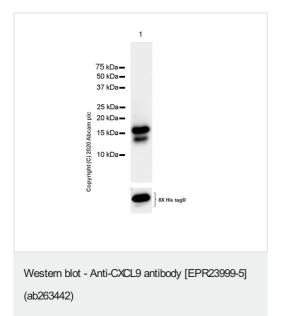
Lane 1: THP-1 treated with 100ng/ml IFN-γ and 1ug/ml LPS (Lipopolysaccharide) for 48 hours, whole cell lysate 10 ug

Lane 2: ab263442 IP in THP-1 treated with 100ng/ml IFN-γ and 1ug/ml LPS(Lipopolysaccharide) for 48 hours whole cell lysate

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab263442 in THP-1 treated with 100ng/ml IFN-γ and 1ug/ml LPS(Lipopolysaccharide) for 48 hours whole cell lysate

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

Exposure time: 3 minutes.



Anti-CXCL9 antibody [EPR23999-5] (ab263442) at 1/1000 dilution + His-tagged human purified recombinant full-length protein CXCL9, 10 ng

Secondary

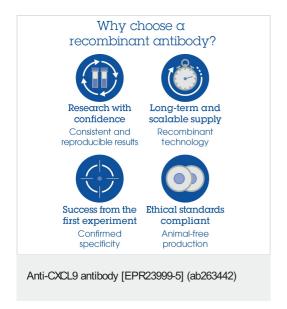
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (<u>ab97051</u>) at 1/50000 dilution

Predicted band size: 14 kDa **Observed band size:** 13.14 kDa

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

The expression profile is consistent with what has been described in the literature (PMID: 15154616, 22973274).

Exposure time: 5.5 seconds.



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