abcam

Product datasheet

Anti-MCP1 antibody [EPR21025] ab214819

KO VALIDATED Recombinant RabMAb

6 Images

Overview

Product name	Anti-MCP1 antibody [EPR21025]
Description	Rabbit monoclonal [EPR21025] to MCP1
Host species	Rabbit
Tested applications	**Suitable for:** ICC, WB, Flow Cyt, IP **Unsuitable for:** IHC-P
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: THP-1 treated with PMA (ab120297), LPS, and BFA whole cell lysate. ICC/IF: THP-1 cells treated with PMA (ab120297), LPS, and BFA, wild-type and MCP1 knockout HeLa cells (ab255372) treated with TNF-alpha (20ng/mL, 6 hours). Flow Cyt: THP-1 treated with PMA (ab120297), LPS, and BFA. IP: THP-1 treated with PMA (ab120297), LPS, and BFA 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	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified

Clonality	Monoclonal
Clone number	EPR21025
Isotype	IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab214819 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

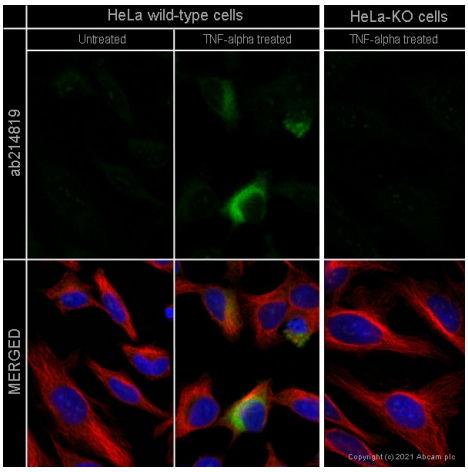
Application	Abreviews	Notes
ICC		1/50.
WB		1/1000. Detects a band of approximately 11 kDa (predicted molecular weight: 11 kDa).
Flow Cyt		1/500.
IP		1/30.

Application notes Is unsuitable for IHC-P.

Target

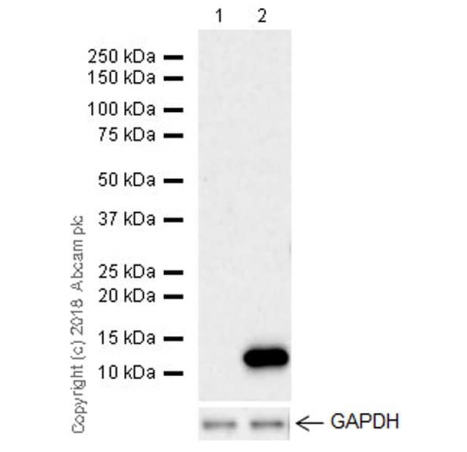
Function	Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils. Augments monocyte anti-tumor activity. Has been implicated in the pathogenesis of diseases characterized by monocytic infiltrates, like psoriasis, rheumatoid arthritis or atherosclerosis. May be involved in the recruitment of monocytes into the arterial wall during the disease process of atherosclerosis.
Sequence similarities	Belongs to the intercrine beta (chemokine CC) family.
Post-translational modifications	Processing at the N-terminus can regulate receptor and target cell selectivity. Deletion of the N-terminal residue converts it from an activator of basophil to an eosinophil chemoattractant.
Cellular localization	Secreted.

Images



Immunocytochemistry - Anti-MCP1 antibody [EPR21025] (ab214819)

ab214819 staining MCP1 in wild-type and MCP1 knockout HeLa cells (ab255372), untreated or treated with TNF-alpha (20ng/ml, 6 hours) and Brefeldin A (1µg/mL, 3 hours). The cells were fixed with 100% methanol (5 min) then permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab214819 at 1µg/ml concentration and ab7291 (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor® 488) (ab150081) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor® 594) (ab150120) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI. Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).



Western blot - Anti-MCP1 antibody [EPR21025] (ab214819)

All lanes : Anti-MCP1 antibody [EPR21025] (ab214819) at 1/1000 dilution

Lane 1 : Untreated THP-1 (human monocytic leukemia cell line) whole cell lysate
Lane 2 : THP-1 treated with 80nM Phorbol-12-myristate-13-acetate (PMA, ab120297) for 24 hours, then treated with 100ng/ml lipopolysaccharide (LPS) for 7 hours, then with 1 µg/ml Brefeldin A (BFA) added after 4 hours, whole cell lysate

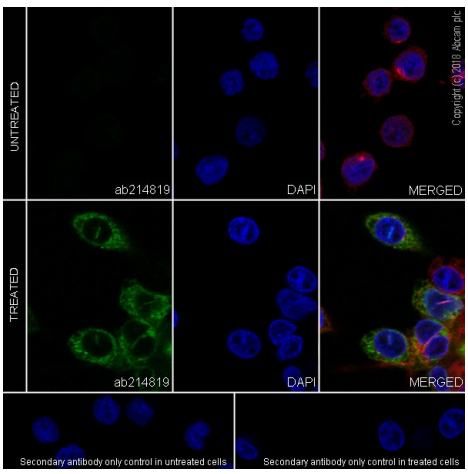
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 11 kDa
Observed band size: 11 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

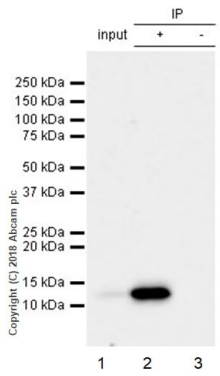


Immunocytochemistry - Anti-MCP1 antibody [EPR21025] (ab214819)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized THP-1 (human monocytic leukemia cell line) cells, untreated or treated with 80nM Phorbol-12-myristate-13-acetate (PMA, ab120297) for 24 hours, then treated with 100ng/ml lipopolysaccharide (LPS) for 7 hours, with 1 µg/ml Brefeldin A (BFA) added after 4 hours, labeling MCP1 with ab214819 at 1/50 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining in THP-1 treated cells.

The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) (red) at 1/200 dilution.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution.



Immunoprecipitation - Anti-MCP1 antibody [EPR21025] (ab214819)

MCP1 was immunoprecipitated from 0.35 mg of THP-1 (human monocytic leukemia cell line) treated with 80nM Phorbol-12-myristate-13-acetate (PMA, ab120297) for 24h, then treated with 100ng/ml lipopolysaccharide (LPS) for 4h, then together with 1µg/ml Brefeldin A (BFA) for another 3h whole cell lysate with ab214819 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab214819 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1,000 dilution

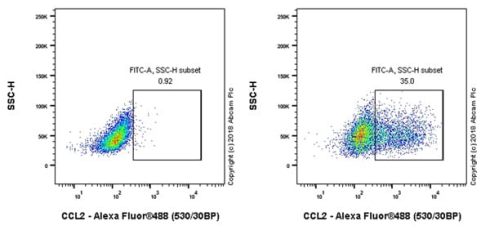
Lane 1: THP-1 treated with 80nM Phorbol-12-myristate-13-acetate (PMA, ab120297) for 24h, then treated with 100ng/ml lipopolysaccharide (LPS) for 4h, then together with 1µg/ml Brefeldin A (BFA) for another 3h whole cell lysate 10 µg (Input).

Lane 2: ab214819 IP in THP-1 treated with 80nM Phorbol-12-myristate-13-acetate (PMA, ab120297) for 24h, then treated with 100ng/ml lipopolysaccharide (LPS) for 4h, then together with 1µg/ml Brefeldin A (BFA) for another 3h whole cell lysate.

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab214819 in THP-1 treated with 80nM Phorbol-12-myristate-13-acetate (PMA,

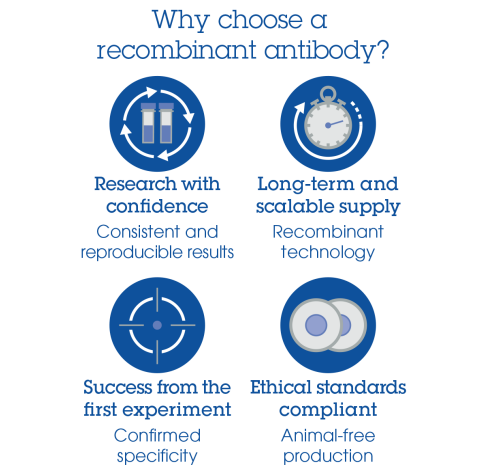
ab120297) for 24h, then treated with 100ng/ml lipopolysaccharide (LPS) for 4h, then together with 1µg/ml Brefeldin A (BFA) for another 3h whole cell lysate.

Blocking and dilution buffer: 5% NFDM/TBST.



Flow Cytometry (Intracellular) - Anti-MCP1 antibody [EPR21025] (ab214819)

Flow cytometric analysis of 4% paraformaldehyde-fixed, 0.1% Tween-20-permeabilized THP-1 (human monocytic leukemia cell line) cell line, treated with 80nM Phorbol-12-myristate-13-acetate (PMA, ab120297) for 24h, then treated with 100ng/ml lipopolysaccharide (LPS) for 4h, then together with 1µg/ml Brefeldin A (BFA) for another 3h (Right) / Untreated control (Left) labeling MCP1 with ab214891 at 1/500 dilution. Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) at 1/2000 dilution was used as the secondary antibody.



Anti-MCP1 antibody [EPR21025] (ab214819)

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