abcam

Product datasheet

Anti-CCR2 antibody [EPR20844-15] ab273050





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Overview

Product name Anti-CCR2 antibody [EPR20844-15]

Description Rabbit monoclonal [EPR20844-15] to CCR2

Host species Rabbit

Tested applications Suitable for: ICC/IF, IHC-P, WB, IP, Flow Cyt

Species reactivity Reacts with: Mouse, Rat

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

Positive control IHC-P: Mouse spleen and lung tissue; Rat spleen tissue. ICC/IF: HEK-293T cells transfected with

> GFP tagged mouse CCR2 overexpression construct. Flow Cyt: C57 BL/6 mouse bone marrow cells, HEK-293T cells transfected with GFP tagged mouse CCR2 overexpression construct. IP:

Mouse lymph node tissue lysate. Mouse bone marrow and spleen.

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, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number EPR20844-15

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab273050 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/IF		1/250.
IHC-P	★★★★☆ (2)	1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Predicted molecular weight: 42 kDa.
IP		1/30.
Flow Cyt		1/600.

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Function Receptor for the MCP-1, MCP-3 and MCP-4 chemokines. Transduces a signal by increasing the

intracellular calcium ions level. Alternative coreceptor with CD4 for HIV-1 infection.

Sequence similarities Belongs to the G-protein coupled receptor 1 family.

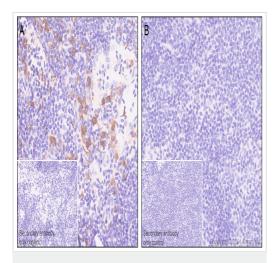
Post-translational

modifications

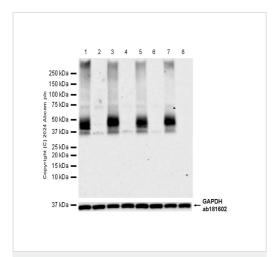
N-glycosylated.

Cellular localization Cell membrane.

Images



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CCR2 antibody
[EPR20844-15] (ab273050)



Western blot - Anti-CCR2 antibody [EPR20844-15] (ab273050)

Immunohistochemical analysis of paraffin-embedded (A) Lymph node tissue from wild-type C57BL/6JGpt mice (B) Lymph node tissue from CCR2 knockout mice staining with ab273050 at 1/300 dilution and ready-to-use Goat Anti-Rabbit IgG H&L (HRP) secondary. Counterstaining with hematoxylin. Heat mediated antigen retrieval was performed with Citrate buffer (pH 6.0, Epitope Retrieval Solution 1) for 20 mins. Positive staining on (A) Lymph node tissue from wild-type C57BL/6JGpt mice and no staining on (B) Lymph node tissue from CCR2 knockout mice. The section was incubated with ab273050 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND™ RX instrument. The tissue samples were kindly provided by GemPharmatech. C57BL/6JGpt wildtype mice and CCR2-KO homozygous mice (Strain ID: T006112).

All lanes : Anti-CCR2 antibody [EPR20844-15] (ab273050) at 1/1000 dilution

Lane 1: Wild-type mouse spleen tissue lysate (male)

Lane 2 : CCR2 knockout mouse spleen tissue lysate (male)

Lane 3: Wild-type mouse lung tissue lysate (male)

Lane 4: CCR2 knockout mouse lung tissue lysate (male)

Lane 5: Wild-type mouse spleen tissue lysate (female)

Lane 6: CCR2 knockout mouse spleen tissue lysate (female)

Lane 7: Wild-type mouse lung tissue lysate (female)

Lane 8: CCR2 knockout mouse lung tissue lysate (female)

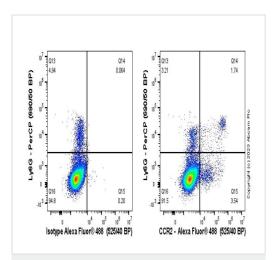
Secondary

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

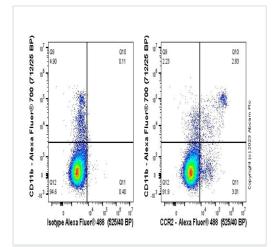
Predicted band size: 42 kDa
Observed band size: 40 kDa

Exposure time: 180 seconds

The tissue samples were kindly provided by GemPharmatech. C57BL/6JGpt wildtype mice and CCR2-KO homozygous mice (Strain ID: T006112).



Flow Cytometry - Anti-CCR2 antibody [EPR20844-15] (ab273050)



Flow Cytometry - Anti-CCR2 antibody [EPR20844-15] (ab273050)

Flow cytometry staining of C57 BL/6 mouse splenocytes with ab273050 (right) or Rabbit monoclonal IgG (ab172730) (left). Cells were incubated for 30 min on ice in 1x PBS containing 10 μ g/ml anti CD16/CD32 and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody ab273050 or Rabbit monoclonal IgG (ab172730) (1x 10⁶ in 100 μ l at 1.0 μ g/ml (1/644)) for 30min on ice. The cells were simultaneously stained with Ly6G.

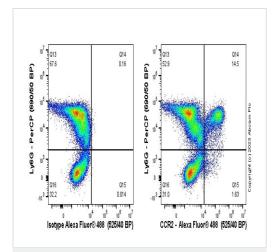
The secondary antibody Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min on ice.

Acquisition of >30000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter. Events were gated on viable cells.

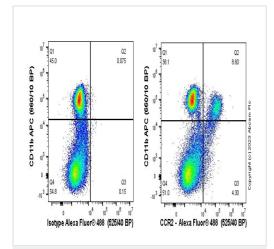
Flow cytometry staining of C57 BL/6 mouse splenocytes with ab273050 (right) or Rabbit monoclonal IgG (ab172730) (left). Cells were incubated for 30 min on ice in 1x PBS containing 10 μ g/ml anti CD16/CD32 and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody ab273050 or Rabbit monoclonal IgG (ab172730) (1x 10⁶ in 100 μ l at 1.0 μ g/ml (1/644)) for 30min on ice. The cells were simultaneously stained with CD11b.

The secondary antibody Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min on ice.

Acquisition of >30000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter. Events were gated on viable cells.



Flow Cytometry - Anti-CCR2 antibody [EPR20844-15] (ab273050)



Flow Cytometry - Anti-CCR2 antibody [EPR20844-15] (ab273050)

Flow cytometry staining of C57 BL/6 mouse bone marrow cells with ab273050 (right) or Rabbit monoclonal IgG (<u>ab172730</u>) (left). Cells were incubated for 30 min on ice in 1x PBS containing 10µg/ml anti CD16/CD32 and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody ab273050 or Rabbit monoclonal IgG (<u>ab172730</u>) (1x 10^6 in 100 µl at 0.2 µg/ml (1/3120)) for 30min on ice. The cells were simultaneously stained with Ly6G.

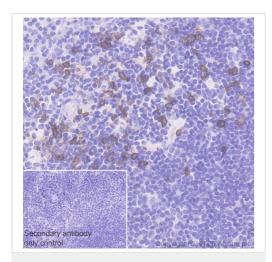
The secondary antibody Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min on ice.

Acquisition of >30000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter. Events were gated on viable cells.

Flow cytometry staining of C57 BL/6 mouse bone marrow cells with ab273050 (right) or Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (left). Cells were incubated for 30 min on ice in 1x PBS containing 10 μ g/ml anti CD16/CD32 and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody ab273050 or Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (1x 10⁶ in 100 μ l at 0.2 μ g/ml (1/12000)) for 30min on ice. The cells were simultaneously stained with CD11b.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min on ice

Acquisition of >30000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter. Events were gated on viable cells.

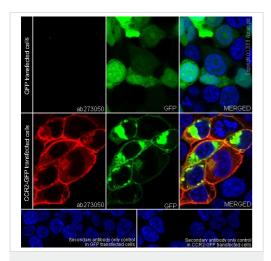


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CCR2 antibody
[EPR20844-15] (ab273050)

Immunohistochemical analysis of paraffin-embedded Mouse spleen tissue labeling CCR2 with ab273050 at 1/250 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on mouse spleen. The section was incubated with ab273050 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

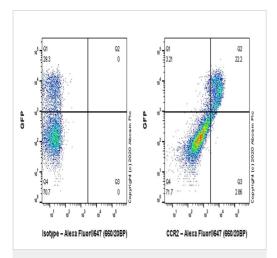
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



Immunocytochemistry/ Immunofluorescence - Anti-CCR2 antibody [EPR20844-15] (ab273050)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HEK-293T (human embryonic kidney epithelial cell) (transfected with GFP tagged mouse CCR2 overexpression construct) cells labelling CCR2 with ab273050 at 1/250 dilution, followed by ab150080 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 594) antibody at 1/1000 dilution (Red). Confocal image showing membranous and cytoplasmic staining in HEK-293T cells transfected with with GFP tagged mouse CCR2 overexpression construct. GFP is shown in Green. The Nuclear counterstain was DAPI (Blue).

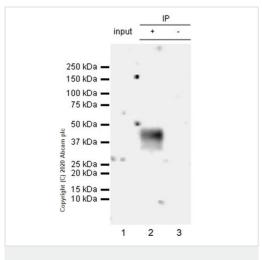
Secondary antibody only control: Secondary antibody is **ab150080** Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 594) at 1/1000 dilution.



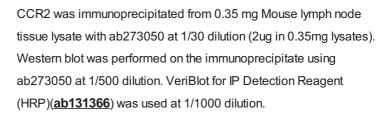
Flow Cytometry - Anti-CCR2 antibody [EPR20844-15] (ab273050)

Flow cytometric analysis of HEK-293T (human embryonic kidney epithelial cell) (transfected with GFP tagged mouse CCR2 overexpression construct) cells labelling CCR2 with ab273050 at 1/600 dilution (Right) compared with a Rabbit monoclonal IgG (ab172730) isotype control (Left). Goat anti rabbit IgG (Alexa Fluor® 647, ab150079) at 1/2000 dilution was used as the secondary antibody.

Gated on viable cells.



Immunoprecipitation - Anti-CCR2 antibody [EPR20844-15] (ab273050)



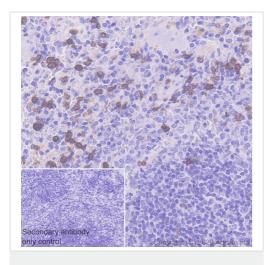
Lane 1: Mouse lymph node tissue lysate 10 ug.

Lane 2: ab273050 IP in Mouse lymph node tissue lysate.

Lane 3:Rabbit monoclonal lgG (<u>ab172730</u>) instead of ab273050 in mouse lymph node tissue lysate.

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

Exposure time: 3 minutes.

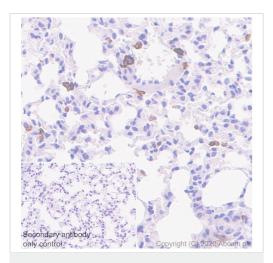


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CCR2 antibody
[EPR20844-15] (ab273050)

Immunohistochemical analysis of paraffin-embedded Rat spleen tissue labeling CCR2 with ab273050 at 1/250 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on rat spleen. The section was incubated with ab273050 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

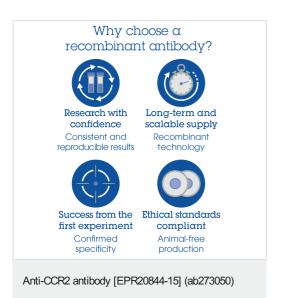


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CCR2 antibody
[EPR20844-15] (ab273050)

Immunohistochemical analysis of paraffin-embedded Mouse lung tissue labeling CCR2 with ab273050 at 1/250 dilutionfollowed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on mouse lung. The section was incubated with ab273050 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



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