CD192 (CCR2) antibodies, human

For research use only

One test corresponds to labeling of up to 10^7 cells in a total volume of 100 µL.

Product	Content	Order no.
CD192 (CCR2)-VioBright FITC	for 30 tests	130-109-658
CD192 (CCR2)-VioBright FITC	for 100 tests	130-109-599
CD192 (CCR2)-PE	for 30 tests	130-109-654
CD192 (CCR2)-PE	for 100 tests	130-109-595
CD192 (CCR2)-APC	for 30 tests	130-109-655
CD192 (CCR2)-APC	for 100 tests	130-109-596
CD192 (CCR2)-PE-Vio770	for 30 tests	130-109-656
CD192 (CCR2)-PE-Vio770	for 100 tests	130-109-597
CD192 (CCR2)-APC-Vio770	for 30 tests	130-109-657
CD192 (CCR2)-APC-Vio770	for 100 tests	130-109-598
CD192 (CCR2)-Biotin	for 30 tests	130-109-653
CD192 (CCR2)-Biotin	for 100 tests	130-109-594

Warnings

Reagents contain sodium azide. Under acidic conditions sodium azide yields hydrazoic acid, which is extremely toxic. Azide compounds should be diluted with running water before discarding. These precautions are recommended to avoid deposits in plumbing where explosive conditions may develop.

Technical data and background information

Antigen	CD192 (CCR2)
Clone	REA624
Isotype	recombinant human IgG1
Isotype control	REA Control (S) antibodies
Alternative names of antigen	C-C chemokine receptor type 2, C-C CKR-2, CC-CKR-2, CCR-2, Monocyte chemoattractant protein 1 receptor, MCP-1-R, MCP-1R
Molecular mass of antigen [kDa]	42
Distribution of antigen	basophils, monocytes
Product format	Reagents are supplied in buffer containing stabilizer and 0.05% sodium azide.
Fixation	The antibody is suited for staining of formaldehyde-fixed cells.
Storage	Store protected from light at 2–8 °C. Do not freeze.

Clone REA624 recognizes the CD192 antigen, a G protein–linked seven transmembrane receptor, which is also known as C-C chemokine receptor type 2 (CCR2) or monocyte chemoattractant protein 1

receptor (MCP-1-R). Two spliced variants of CD192 (CD192A and CD192B) are expressed on peripheral monocytes and basophils as a result of alternate splicing of a single gene and differ at the C-terminal end. CD192 and its main ligand CCL2 have been implicated in a wide range of immunobiological processes and neuropathologies, including recruitment of monocytes and regulation of bone marrow homeostasis, as well as multiple sclerosis, HIV-associated dementia, Alzheimer's disease, and neuropathic pain.
Additional information: Clone REA624 displays negligible binding to Fc receptors.

Reagent requirements

- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS® BSA Stock Solution (# 130-091-376) 1:20 with autoMACS® Rinsing Solution (# 130-091-222). Keep buffer cold (2−8 °C).
  Note: EDTA can be replaced by other supplements such as anticoagulant citrate dextrose formula-A (ACD-A) or citrate phosphate dextrose (CPD). Buffers or media containing Ca^{2+} or Mg^{2+} are not recommended for use.
- (Optional) Fluorochrome-conjugated anti-biotin antibodies, e.g., Anti-Biotin-PE (# 130-090-756) as secondary antibody reagent in combination with biotinylated antibodies.
- (Optional) Propidium Iodide Solution (# 130-093-233) for flow cytometric exclusion of dead cells without fixation.
- (Optional) Fixation and Dead Cell Discrimination Kit (# 130-091-163) for cell fixation and flow cytometric exclusion of dead cells.

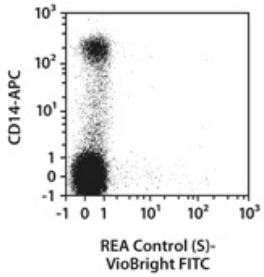
Protocol for cell surface staining

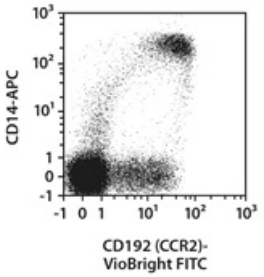
- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:11 for up to 10^7 cells/100 µL of buffer.
- Volumes given below are for up to 10^7 nucleated cells. When working with fewer than 10^7 cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes accordingly (e.g. for 2×10^7 nucleated cells, use twice the volume of all indicated reagent volumes and total volumes).

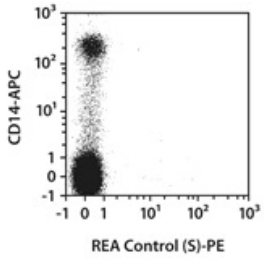
1. Determine cell number.
2. Centrifuge cell suspension at 300×g for 10 minutes. Aspirate supernatant completely.
3. Resuspend up to 10^7 nucleated cells per 100 µL of buffer.
4. Add 10 µL of the antibody.
5. Mix well and incubate for 10 minutes in the dark in the refrigerator (2−8 °C).
   Note: Higher temperatures and/or longer incubation times may lead to non-specific cell labeling. Working on ice requires increased incubation times.
6. Wash cells by adding 1−2 mL of buffer and centrifuge at 300×g for 10 minutes. Aspirate supernatant completely.
7. (Optional) If biotinylated antibody was used, resuspend the cell pellet in 100 µL of buffer, add 10 µL of fluorochrome-conjugated anti-biotin antibody, and continue as described in steps 5 and 6.
8. Resuspend cell pellet in a suitable amount of buffer for analysis by flow cytometry or fluorescence microscopy.

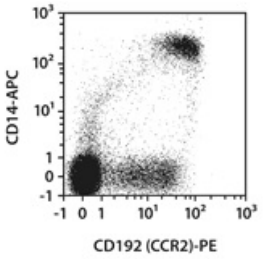
Examples of immunofluorescent staining

Human peripheral blood mononuclear cells (PBMCs) were stained with CD192 (CCR2) antibodies or with the corresponding REA Control (S) antibodies (left image) as well as with CD14 antibodies. Flow cytometry was performed using the MACSQuant® Analyzer. The Tandem Signal Enhancer has been used to increase binding specificity of tandem-dye–conjugated antibodies. Cell debris and dead cells were excluded from the analysis based on scatter signals and propidium iodide fluorescence or 4',6-diamidino-2-phenylindole (DAPI) fluorescence, as in the case of tandem conjugates.


CD14-APC
REA Control (S)-
VioBright FITC


CD14-APC
CD192 (CCR2)-
VioBright FITC


CD14-APC
REA Control (S)-PE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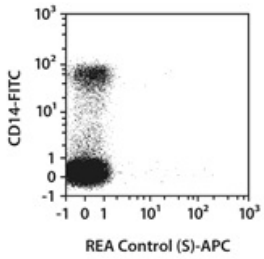
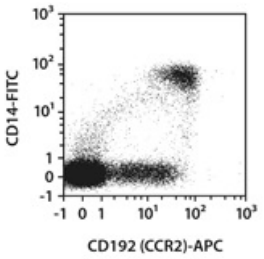

CD14-FITC
REA Control (S)-APC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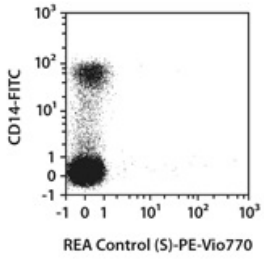
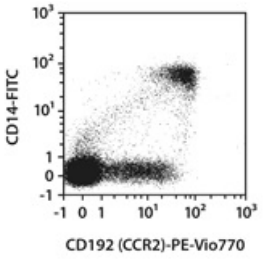

CD14-FITC
REA Control (S)-PE-Vio770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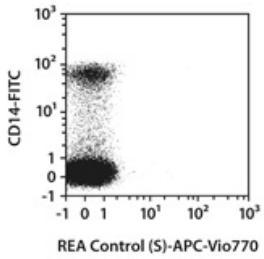
CD192 (CCR2)-PE-Vio770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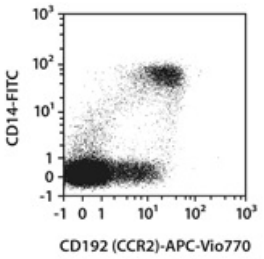
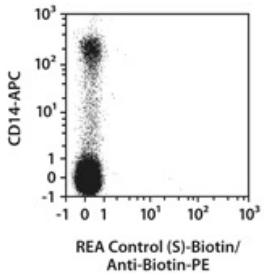
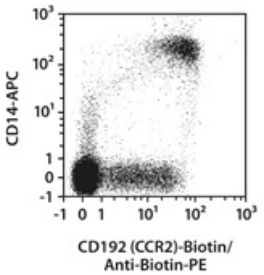
REA Control (S)-APC-Vio770


CD14-FITC
CD192 (CCR2)-APC-Vio770


CD14-APC
REA Control (S)-Biotin/
Anti-Biotin-PE


CD14-APC
CD192 (CCR2)-Biotin/
Anti-Biotin-PE

References

1. **Charo, I. F. *et al.*** (1994) Molecular cloning and functional expression of two monocyte chemoattractant protein 1 receptors reveals alternative splicing of the carboxyl-terminal tails. Proc. Natl. Acad. Sci. U.S.A. 91(7): 2752–2756.
2. **Wong, L. M. *et al.*** (1997) Organization and differential expression of the human monocyte chemoattractant protein 1 receptor gene. Evidence for the role of the carboxyl-terminal tail in receptor trafficking. J. Biol. Chem. 272(2): 1038–1045.
3. **Henrich T. J. *et al.*** (2013) HIV-1 entry inhibitors: recent development and clinical use. Curr. Opin. Virol. 3(1): 51–57.

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