CD301a (MGL1) antibodies, mouse

For research use only

9 µg equal 60 tests, 30 µg equal 200 tests. One test corresponds to labeling of 10^6 cells.

Product	Content	Order no.
CD301a (MGL1)-VioBright FITC	9 µg in 300 µL	130-109-297
CD301a (MGL1)-VioBright FITC	30 µg in 1 mL	130-109-219
CD301a (MGL1)-PE	9 µg in 300 µL	130-109-294
CD301a (MGL1)-PE	30 µg in 1 mL	130-109-216
CD301a (MGL1)-APC	9 µg in 300 µL	130-109-295
CD301a (MGL1)-APC	30 µg in 1 mL	130-109-217
CD301a (MGL1)-PE-Vio770	9 µg in 300 µL	130-109-296
CD301a (MGL1)-PE-Vio770	30 µg in 1 mL	130-109-218
CD301a (MGL1)-Biotin	9 µg in 300 µL	130-109-293
CD301a (MGL1)-Biotin	30 µg in 1 mL	130-109-215

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	CD301a (MGL1)
Clone	REA581
Isotype	recombinant human IgG1
Isotype control	REA Control antibodies
Alternative names of antigen	Clec10a, MMGL, M-ASGP-BP-1
Molecular mass of antigen [kDa]	35
Distribution of antigen	macrophages
Product format	Reagents are supplied in buffer containing stabilizer and 0.05% sodium azide.
Fixation	Cells should be stained prior to fixation, if formaldehyde is used as a fixative.
Storage	Store protected from light at 2–8 °C. Do not freeze.

Clone REA581 recognizes the mouse CD301a antigen, a type II transmembrane glycoprotein also known as macrophage galactose-type C-type lectin-1 (MGL1). CD301 has two genes, CD301a and CD301b, which are highly homologous. In contrast to CD301b, that recognizes N-acetylgalactosamine and galactose, including the O-linked Tn antigen, TF antigen, and core 2, CD301a is highly specific for Lewis X and Lewis A structures. It has been suggested that MGL1 is involved in the clearance of

apoptotic cells. CD301a is expressed in alternatively activated macrophages.
Additional information: Clone REA581 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.

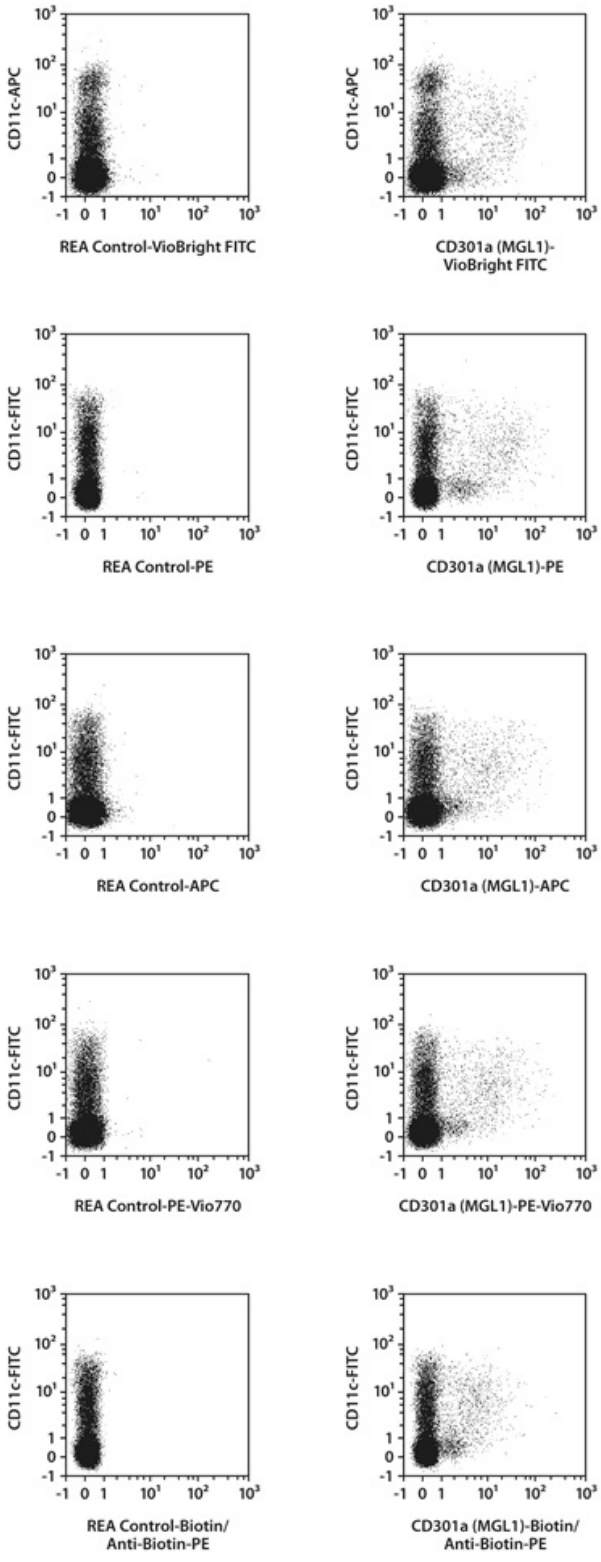
Protocol for cell surface staining

- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:10 for up to 10^6 cells/50 µL of buffer.
- Volumes given below are for up to 10^6 nucleated cells. When working with fewer than 10^6 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 2×10^6 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 10^6 nucleated cells per 45 µL of buffer.
4. Add 5 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

Splenocytes from C57BL/6 mice were stained with CD301a (MGL1) antibodies or with the corresponding REA Control antibodies (left image) as well as with CD11c antibodies. $CD3^-/CD19^-$ cells were pregated for the analysis. Flow cytometry was performed using the MACSQuant® Analyzer. Cell debris and dead cells were excluded from the analysis based on scatter signals and propidium iodide fluorescence.



References

1. **Raes, G. *et al.*** (2005) Macrophage galactose-type C-type lectins as novel markers for alternatively activated macrophages elicited by parasitic infections and allergic airway inflammation. J. Leukoc. Biol. 77(3): 321–327.
2. **Yuita, H. *et al.*** (2005) Retardation of removal of radiation-induced apoptotic cells in developing neural tubes in macrophage galactose-type C-type lectin-1-deficient mouse embryos. Glycobiology 15(12): 1368–1375.
3. **Singh, S. K. *et al.*** (2009) Characterization of murine MGL1 and MGL2 C-type lectins: distinct glycan specificities and tumor binding properties. Mol. Immunol. 46(6): 1240–1249.

Warranty

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