

# CD205 (DEC205) antibodies, mouse

## For research use only

30 μg equal 100 tests, 150 μg equal 500 tests. One test corresponds to labeling of  $10^6$  cells.

Product	Content	Order no.
CD205 (DEC205)-Biotin	150 μg in 1 mL	130-112-111
CD205 (DEC205)-PE	30 μg in 200 μL	130-112-272
CD205 (DEC205)-PE	150 μg in 1 mL	130-112-114
CD205 (DEC205)-APC	30 μg in 200 μL	130-112-273
CD205 (DEC205)-APC	150 μg in 1 mL	130-112-115
CD205 (DEC205)-PE-Vio615	30 μg in 200 μL	130-112-270
CD205 (DEC205)-PE-Vio615	150 μg in 1 mL	130-112-112
CD205 (DEC205)-PE-Vio770	30 μg in 200 μL	130-112-274
CD205 (DEC205)-PE-Vio770	150 μg in 1 mL	130-112-116
CD205 (DEC205)-Biotin	30 μg in 200 μL	130-112-269

# **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 CD205 (DEC205)

Clone REA817

Isotyperecombinant human IgG1Isotype controlREA Control antibodiesAlternative names of antigenLY75, DEC-205, DEC205

Entrez Gene ID 17076

Molecular mass of antigen [kDa] 195

**Distribution of antigen** dendritic cells, skin, Langerhans cells, T cells

**Product format** Reagents are supplied in buffer containing stabilizer and 0.05% sodium azide.

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**Fixation** The antibody is suited for staining of formaldehyde-fixed cells.

**Storage** Store protected from light at 2–8 °C. Do not freeze.

Clone REA817 recognizes the mouse CD205 antigen, a 205 kDa integral membrane glycoprotein also known as DEC205 (dendritic and epithelial cells, 205 kDa). This membrane protein acts as an endocytic receptor and thereby mediates efficient processing and presentation of antigens in vivo, leading to the induction of T cell immunity or tolerance. CD205 is expressed at high levels on mouse dendritic cells (DCs) in the skin (Langerhans cells), on DCs residing in the T cell areas of peripheral lymphoid organs, and on DCs generated in vitro from bone marrow progenitors. To a much lower extent, CD205 is also expressed on mature B cells, granulocytes, and T cells.

#### **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<sup>2+</sup> or Mg<sup>2+</sup> 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.

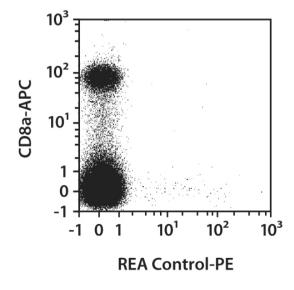
## **Protocol for cell surface staining**

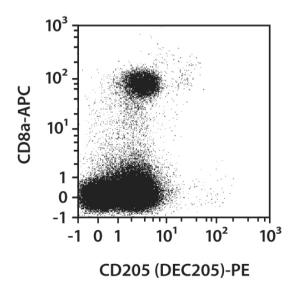
- $^{\circ}$  The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:50 for up to  $10^{^{\circ}}$  cells/100  $\mu$ L.
- $^{\bullet}$  Volumes given below are for up to  $10^{^{\circ}}$  nucleated cells. When working with fewer than  $10^{^{\circ}}$  cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes accordingly.
- 1. Determine cell number.
- 2. Centrifuge cell suspension at 300×g for 10 minutes. Aspirate supernatant completely.
- 3. Resuspend up to  $10^{\circ}$  nucleated cells per 98 µL of buffer.
- 4. Add 2  $\mu L$  of the antibody.
- 5. Mix well and incubate for 10 minutes in the dark in the refrigerator (2–8 °C).

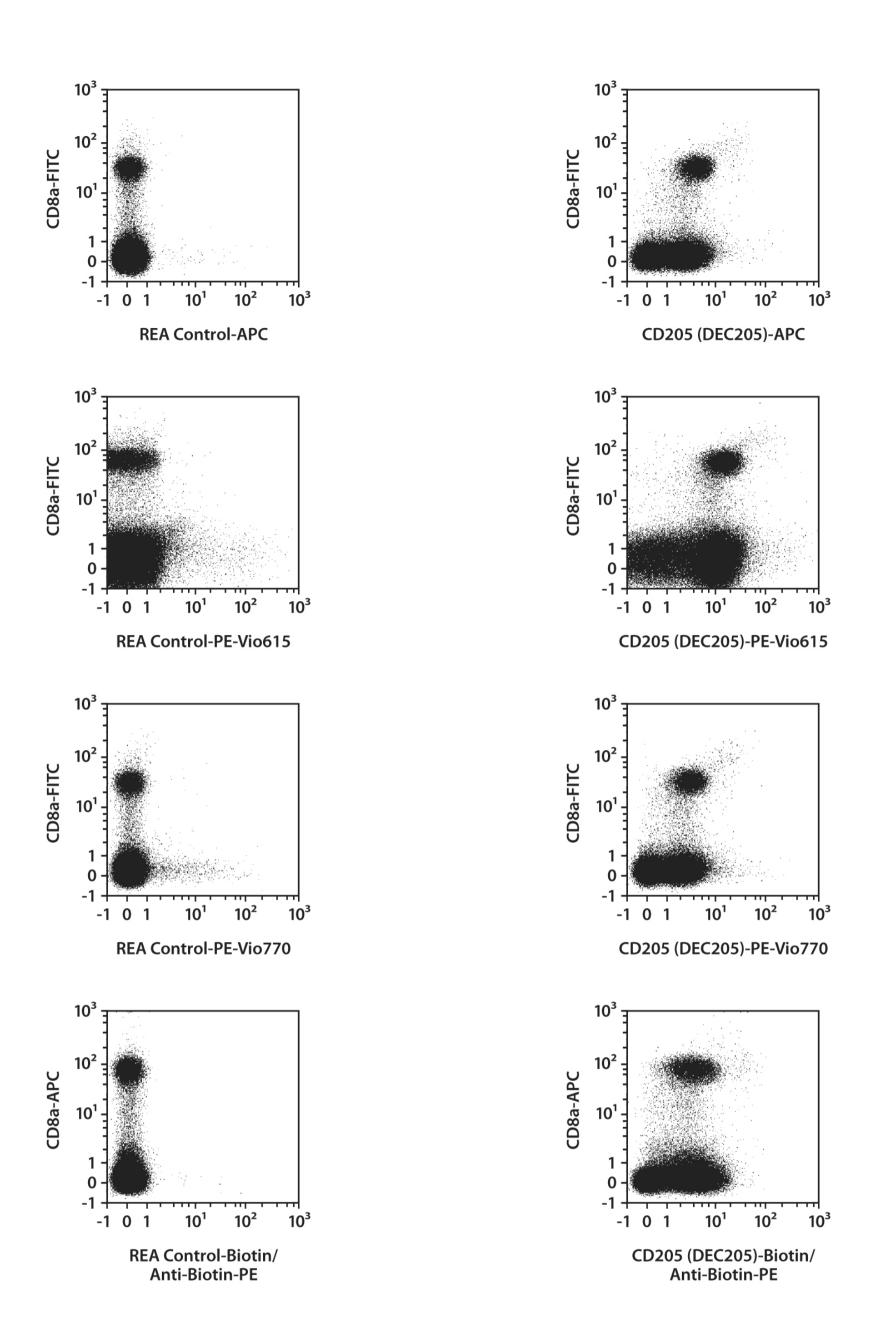
  Note: Higher temperatures and/or longer incubation times may lead to non-specific cell labeling. Working on ice requires increased incubation times.
- 6. Wash cells by adding 1-2 mL of buffer and centrifuge at  $300\times g$  for 10 minutes. Aspirate supernatant completely.
- 7. (Optional) If biotinylated antibody was used, resuspend the cell pellet in buffer and stain with fluorochrome-conjugated antibiotin antibody according to the manufacturer's recommendations.
- 8. Resuspend cell pellet in a suitable amount of buffer for analysis by flow cytometry or fluorescence microscopy.

### **Examples of immunofluorescent staining**

Splenocytes of C57BL/6 mice were stained with CD205 (DEC205) antibodies or the corresponding REA Control antibodies (left image) as well as with CD8a antibodies. Flow cytometry was performed using the MACSQuant<sub>®</sub>Analyzer. 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.







## Warranty

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