

# CD133/1 (AC133) antibodies, human

## For research use only

One test corresponds to labeling of up to  $10^{\circ}$  cells in a total volume of  $100~\mu L$ 

Product	Content	Order no.
CD133/1 (AC133)-APC	for 30 tests	130-113-668
CD133/1 (AC133)-VioBright FITC	for 30 tests	130-113-673
CD133/1 (AC133)-VioBright FITC	for 100 tests	130-113-111
CD133/1 (AC133)-PE	for 30 tests	130-113-670
CD133/1 (AC133)-PE	for 100 tests	130-113-108
CD133/1 (AC133)-APC	for 100 tests	130-113-106
CD133/1 (AC133)-PE-Vio615	for 30 tests	130-113-671
CD133/1 (AC133)-PE-Vio615	for 100 tests	130-113-109
CD133/1 (AC133)-PE-Vio770	for 30 tests	130-113-672
CD133/1 (AC133)-PE-Vio770	for 100 tests	130-113-110
CD133/1 (AC133)-VioBright 515	for 30 tests	130-113-674
CD133/1 (AC133)-VioBright 515	for 100 tests	130-113-112
CD133/1 (AC133)-Biotin	for 30 tests	130-113-669
CD133/1 (AC133)-Biotin	for 100 tests	130-113-107

## **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 CD133/1 (AC133)

Clone AC133

**Isotype** mouse IgG1κ

**Isotype control** Mouse IgG1 – isotype control antibodies

Alternative names of antigen PROM1, AC133, CORD12, MCDR2, MSTP061, PROML1, RP41, STGD4

Entrez Gene ID 8842
Molecular mass of antigen [kDa] 95

**Distribution of antigen** brain, endothelial cells, epithelial cells, heart, hematopoietic stem cells, kidney, liver,

lung, pancreas, placenta, ES and iPS cells, red blood cells

**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.

CD133, formerly known as AC133, recognizes epitope 1 of the CD133 antigen. It is a marker that is frequently found on multipotent progenitor cells, including immature hematopoietic stem and progenitor cells. In the hematopoietic system, CD133 is expressed on a small portion of CD34 cells as well as on a subset of CD34 stem and progenitor cells in human fetal liver, bone marrow, cord blood, and peripheral blood. CD133 has also been found to be expressed on circulating endothelial progenitor cells, fetal neural stem cells, other tissue-specific stem cells, such as renal, prostate, and corneal stem cells, cancer stem cells from tumor tissues, as well as ES and iPS cell-derived 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) FcR Blocking Reagent, human (# 130-059-901) to avoid Fc receptor-mediated antibody labeling.
- (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.

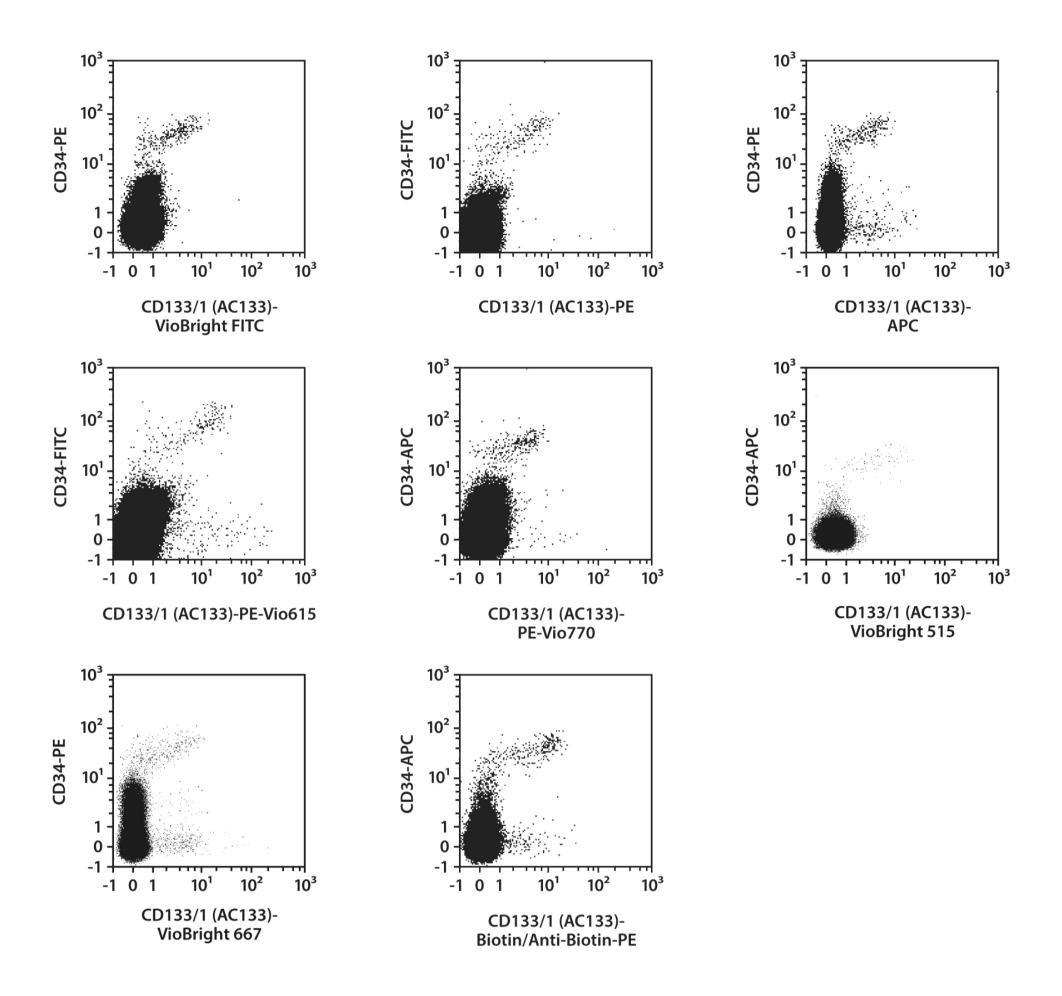
## **Protocol for cell surface staining**

- $^{\circ}$  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  $10^6$  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**

Human peripheral blood mononuclear cells (PBMCs) were stained with CD133/1 (AC133) antibodies as well es with CD34 and CD45 antibodies and analyzed by flow cytometry using the MACSQuant®Analyzer. The Tandem Signal Enhancer has been used to increase binding specificity of tandem-dye-conjugated antibodies. For all other conjugates the FcR Blocking Reagent has been used to avoid Fc receptor-mediated antibody labeling. A pre-gate of CD45+cells was used. 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 tandems.



#### **Warranty**

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