

# CD133/1 (AC133) antibodies, human

For research use only

One test corresponds to labeling of up to  $10^7$  cells in a total volume of 100  $\mu$ L.

Product	Content	Order no.
CD133/1 (AC133)-VioBright FITC <sup>1</sup>	for 30 tests	130-105-226
CD133/1 (AC133)-VioBright FITC <sup>1</sup>	for 100 tests	130-105-225
CD133/1 (AC133)-PE <sup>1</sup>	for 30 tests	130-098-826
CD133/1 (AC133)-PE <sup>1</sup>	for 100 tests	130-080-801
CD133/1 (AC133)-APC <sup>1</sup>	for 30 tests	130-098-829
CD133/1 (AC133)-APC <sup>1</sup>	for 100 tests	130-090-826
CD133/1-PE-Vio615 <sup>1</sup>	for 30 tests	130-107-509
CD133/1-PE-Vio615 <sup>1</sup>	for 100 tests	130-107-455
CD133/1-PE-Vio770 <sup>1</sup>	for 30 tests	130-101-652
CD133/1-PE-Vio770 <sup>1</sup>	for 100 tests	130-101-676
CD133/1 (AC133)-Biotin <sup>1</sup>	for 30 tests	130-098-897
CD133/1 (AC133)-Biotin <sup>1</sup>	for 100 tests	130-090-664
CD133/1 (AC133) pure	50 $\mu$ g in 1 mL	130-090-422

<sup>1</sup>Not recommended for cells that are labeled with MACS MicroBeads using the same antigen.

## 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

<b>Antigen</b>	CD133/1 (AC133)
<b>Clone</b>	AC133
<b>Isotype</b>	mouse IgG1 $\kappa$
<b>Isotype control</b>	Mouse IgG1 – isotype control antibodies
<b>Alternative names of antigen</b>	PROM1, AC133, CORD12, MCDR2, MSTP061, PROML1, RP41, STGD4
<b>Molecular mass of antigen [kDa]</b>	95
<b>Distribution of antigen</b>	brain, endothelial cells, epithelial cells, heart, hematopoietic stem cells, kidney, liver, lung, pancreas, placenta, ES and iPS cells, red blood cells
<b>Product format</b>	Antibodies are supplied in buffer containing stabilizer and 0.05% sodium azide.

<b>Fixation</b>	Cells should be stained prior to fixation, if formaldehyde is used as a fixative.
<b>Storage</b>	Store protected from light at 2–8 °C. Do not freeze.

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<sup>-</sup> cells as well as on a subset of CD34<sup>bright</sup> 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

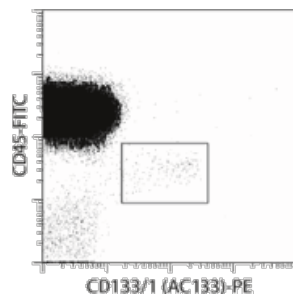
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS<sup>®</sup> BSA Stock Solution (# 130-091-376) 1:20 with autoMACS<sup>®</sup> Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).  
Note: EDTA can 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

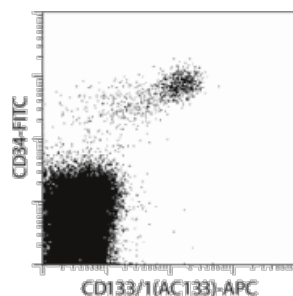
- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:11 for up to 10<sup>7</sup> cells/100 µL of buffer.
  - Volumes given below are for up to 10<sup>7</sup> nucleated cells. When working with fewer than 10<sup>7</sup> cells, use the same volumes as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

Staining of non-mobilized PBMCs with CD133/1-PE and CD45-FITC.



Human peripheral blood mononuclear cells (PBMCs) were stained with CD133/1 (AC133) antibodies conjugated to APC, as well as with CD34-FITC. The FcR Blocking Reagent has been used to avoid Fc receptor-mediated antibody labeling. Cells were analyzed by flow cytometry. Cell debris and dead cells were excluded from the analysis based on scatter signals and propidium iodide fluorescence.



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