CD34 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.
CD34-FITC	for 30 tests	130-098-142
CD34-FITC	for 100 tests	130-081-001
CD34-PE	for 30 tests	130-098-140
CD34-PE	for 100 tests	130-081-002
CD34-APC	for 30 tests	130-098-139
CD34-APC	for 100 tests	130-090-954
CD34-VioBlue	for 30 tests	130-098-144
CD34-VioBlue	for 100 tests	130-095-393
CD34-VioGreen	for 30 tests	130-106-844
CD34-VioGreen	for 100 tests	130-106-800
CD34-PE-Vio770	for 30 tests	130-100-844
CD34-PE-Vio770	for 100 tests	130-096-740
CD34-PerCP-Vio700	for 100 tests	130-097-915
CD34-Biotin	for 30 tests	130-098-553
CD34-Biotin	for 100 tests	130-098-554

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	CD34
Clone	AC136
Isotype	mouse IgG2a
Isotype control	Mouse IgG2a – isotype control antibodies
Alternative names of antigen	gp105-120, Mucosialin, My10
Molecular mass of antigen [kDa]	37
Distribution of antigen	bone marrow, cancer stem cells, CNS cells, dendritic cells, endothelial cells, fibroblasts, hematopoietic stem cells, liver, mast cells, osteoblasts
Product format	Antibodies 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.

The monoclonal antibody clone AC136 detects a class III epitope of the CD34 antigen. This epitope is different than the one recognized by the clone used in the CD34 MicroBead Kits.
The CD34 antigen is a single chain transmembrane glycoprotein, expressed on human hematopoitetic stem and progenitor cells, endothelial progenitor cells, vascular endothelial cells, embryonic fibroblasts, and some cells in fetal and adult nervous tissue. The antigen is absent on fully differentiated hematopoietic cells such as normal peripheral blood lymphocytes, monocytes, granulocytes, erythrocytes, and platelets. Clone AC136 has a similar specifity as the CD34 monoclonal antibody clone 8G12 (HPCA-2).
CD34 antibodies can be used for studies of hematopoiesis and nonhematopoiectic stem cells, phenotyping of hematopoietic stem cells, and studies on phenotyping of hematologic malignancies, endothelial cells, and endothelial progenitor cells (EPCs).

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) 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

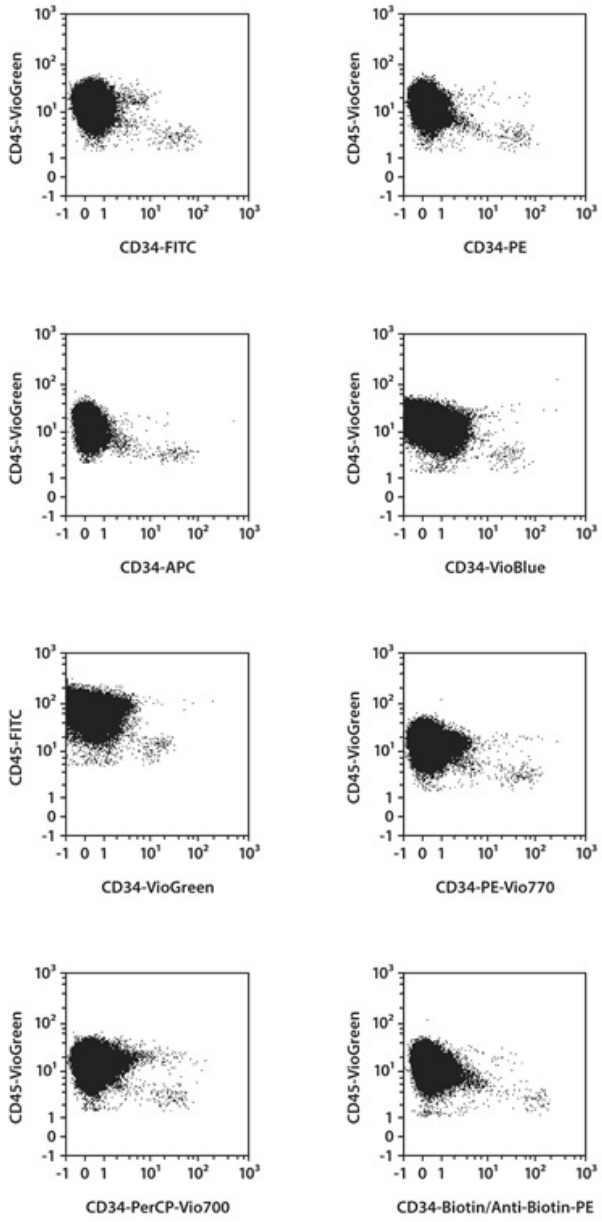
- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:11 for up to 10^7 cells/100 µL of buffer.
- Volumes given below are for up to 10^7 nucleated cells. When working with fewer than 10^7 cells, use the same volumes as indicated. When working with higher 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 CD34 antibodies as well as with CD45 and CD15 antibodies. 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. The cells were analyzed by flow cytometry using the MACSQuant® Analyzer. A pregate of CD45+/CD15- 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.



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Warranty

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