

CD15 antibodies, human

For research use only

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

Product	Content	Order no.
CD15-FITC	for 30 tests	130-098-013
CD15-FITC	for 100 tests	130-081-101
CD15-PE	for 30 tests	130-098-010
CD15-PE	for 100 tests	130-091-375
CD15-APC	for 30 tests	130-098-008
CD15-APC	for 100 tests	130-091-371
CD15-VioBlue	for 30 tests	130-099-079
CD15-VioBlue	for 100 tests	130-099-078
CD15-VioGreen	for 30 tests	130-098-016
CD15-VioGreen	for 100 tests	130-096-877
CD15-PE-Vio770	for 30 tests	130-100-426
CD15-PE-Vio770	for 100 tests	130-100-425
CD15-PerCP-Vio700	for 30 tests	130-100-428
CD15-PerCP-Vio700	for 100 tests	130-100-462
CD15-Biotin	for 100 tests	130-098-383

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 CD15
Clone VIMC6
Isotype mouse IgM

Isotype control Mouse IgM – isotype control antibodies

Alternative names of antigen FUT4, ELFT, FCT3A, FUC-TIV, FUTIV, Lex, SSEA-1, Lewis X

Distribution of antigen B cells, basophils, bone marrow, brain, breast, cancer stem cells,

eosinophils, epithelial cells, granulocytes, kidney, Langerhans cells, leukemia cells, lung, monocytes, myeloid cells, neutrophils, T cells

Product format Antibodies are supplied in buffer containing stabilizer and 0.05%

sodium azide.

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

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

The CD15 antibody recognizes the CD15 antigen which is expressed on human myelomonocytic cells. It is present on neutrophils, eosinophils, and some monocytes, but not on basophils or lymphocytes. The antigen is also expressed on Hodgkin/Reed-Sternberg cells.

The CD15 antibody recognizes the carbohydrate structure 3-fucosyl-N-acetyl-lactosamine. This structure is also known as stage specific embryonic antigen 1 (SSEA-1) and a common surface marker for pluripotency on mouse ES and iPS cells. On human cells in contrast, SSEA-1 is expressed only on early differentiating cells. The CD15 antibody recognizes the SSEA-1 antigen in both species. For applications in ES and iPS research, the dilution recommended in the data sheet may need adjustment.

Reagent requirements

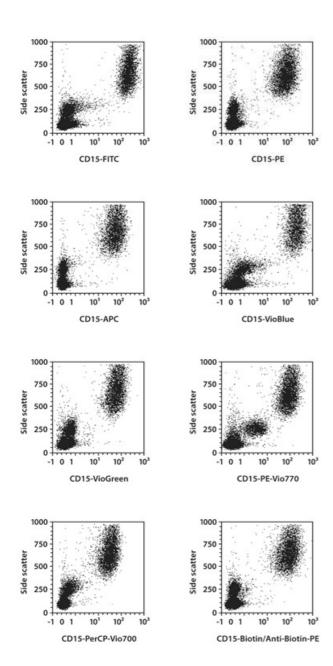
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS[®] BSA Stock Solution (# 130-091-376) 1:20 with autoMACS[®] Rinsing Solution (# 130-091-222). Keep buffer cold (2-8 °C). Note: EDTA can be replaced by other supplements such as anticoagulant citrate dextrose formula-A (ACD-A) or citrate phosphate dextrose (CPD). Buffers or media containing Ca²⁺ or Mg²⁺ 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 10⁷ cells/100 µL of buffer.
- Volumes given below are for up to 10⁷ nucleated cells. When working with fewer than 10⁷ 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 2×10⁷ 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 μ L of buffer.
- 4. Add 10 µL of the antibody.
- 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 μ L of buffer, add 10 μ L 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

White blood cells (WBCs) were stained with CD15 antibodies and analyzed by flow cytometry. 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.



References

 Choi, K.-D. et al. (2011) Hematopoietic differentiation and production of mature myeloid cells from human pluripotent stem cells. Nat. Protoc. 6(3): 296–313.

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