CD44 antibodies, mouse

For research use only

9 µg equal 60 tests, 30 µg equal 200 tests. One test corresponds to labeling of 10^6 cells.

Product	Content	Order no.
CD44-FITC	9 µg in 300 µL	130-102-933
CD44-FITC	30 µg in 1 mL	130-102-511
CD44-PE	9 µg in 300 µL	130-102-982
CD44-PE	30 µg in 1 mL	130-102-606
CD44-APC	9 µg in 300 µL	130-102-977
CD44-APC	30 µg in 1 mL	130-102-563
CD44-VioBlue	9 µg in 300 µL	130-102-976
CD44-VioBlue	30 µg in 1 mL	130-102-443
CD44-PE-Vio770	9 µg in 300 µL	130-102-904
CD44-PE-Vio770	30 µg in 1 mL	130-102-377
CD44-APC-Vio770	9 µg in 300 µL	130-102-965
CD44-APC-Vio770	30 µg in 1 mL	130-102-326
CD44-Biotin	9 µg in 300 µL	130-102-020
CD44-Biotin	30 µg in 1 mL	130-101-946

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	CD44
Clone	IM7.8.1
Isotype	rat IgG2bκ
Isotype control	Rat IgG2b – isotype control antibodies
Alternative names of antigen	HERMES, Ly-24, Pgp-1, CD44s, EMCR III, H-CAM
Molecular mass of antigen [kDa]	83
Cross-reactivity	rhesus monkey (*Macaca mulatta*), cynomolgus monkey (*Macaca fascicularis*), chimpanzee (*Pan troglodytes*), baboon, human, pig, cow, horse, cat, dog
Distribution of antigen	bone marrow, cancer stem cells, CNS cells, endothelial cells, epithelial cells, kidney, leukocytes, lymphocytes, mesenchymal stem cells, myeloid cells, plasma cells, ES and iPS cells, red blood cells, skeletal muscle, skin, 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.

Mouse CD44 is also known as Pgp-1, HCELL, HUTCH-I, HCAM, HERMES, and Ly-24. Clone IM7 reacts with murine CD44, an 80–95 kDa membrane glycoprotein. It is expressed by a variety of hematopoetic and nonhematopoetic cells. Different isoforms are generated by alternative splicing. Clone IM7 reacts with all isoforms and both allelic forms of CD44. CD44 functions as a receptor for hyaluronic acid (HA) and mediates cell-cell and cell-matrix interactions through its affinity for HA, and possibly also through its affinity for other ligands, such as osteopontin, collagens, and matrix metalloproteinases (MMPs). CD44 expression is upregulated on naive T cells upon activation and high CD44 expression is maintained on memory T cells. Therefore, CD44 is often used to discriminate $CD44^{hi}$ antigen-experienced T cells from $CD44^{lo}$ ($CD62L^{+}$) naive 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^{2+} or Mg^{2+} are not recommended for use.
- (Optional) FcR Blocking Reagent, mouse (# 130-092-575) 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.

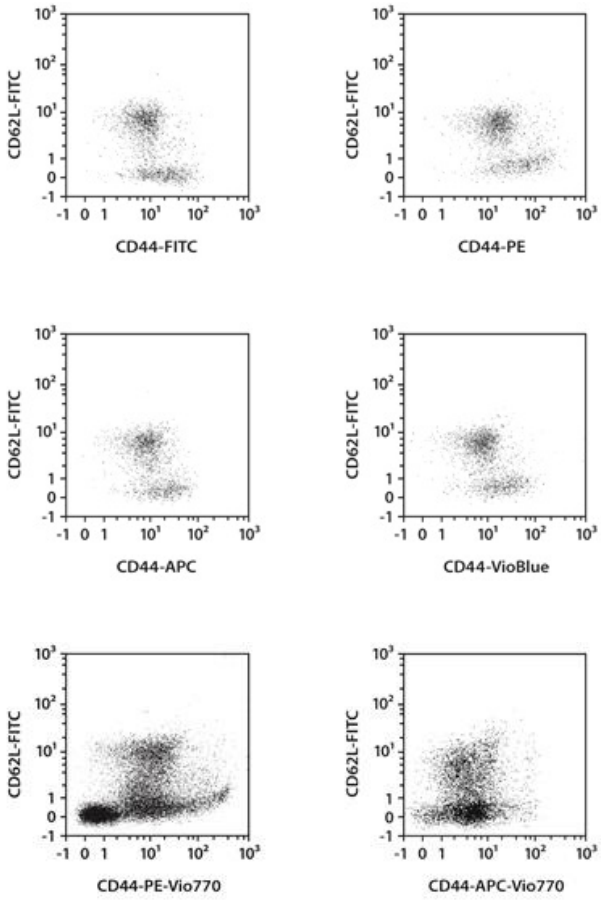
Protocol for cell surface staining

- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:10 for up to 10^6 cells/50 µL of buffer.
- Volumes given below are for up to 10^6 nucleated cells. When working with fewer than 10^6 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 2×10^6 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 10^6 nucleated cells per 45 µL of buffer.
4. Add 5 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

Balb/c spleen cells were stained with CD44 antibodies. Cells were analyzed by flow cytometry using the MACSQuant® Analyzer and 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 CD44-PE-Vio770 and CD44-APC-Vio770.



References

1. **Ponta, H. *et al.*** (2003) CD44: From adhesion molecules to signalling regulators. Nat. Rev. Mol. Cell Biol. 4(1): 33–45.

Warranty

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