

CD44 antibodies, human

For research use only

One test corresponds to labeling of up to 10° cells in a total volume of $100~\mu L$

Product	Content	Order no.
CD44-APC-Vio770	for 100 tests	130-113-332
CD44-FITC	for 30 tests	130-113-896
CD44-FITC	for 100 tests	130-113-334
CD44-PE	for 30 tests	130-113-897
CD44-PE	for 100 tests	130-113-335
CD44-APC	for 30 tests	130-113-893
CD44-APC	for 100 tests	130-113-331
CD44-VioBlue	for 30 tests	130-113-899
CD44-VioBlue	for 100 tests	130-113-337
CD44-PE-Vio770	for 30 tests	130-113-898
CD44-PE-Vio770	for 100 tests	130-113-336
CD44-APC-Vio770	for 30 tests	130-113-894
CD44-Biotin	for 30 tests	130-113-895
CD44-Biotin	for 100 tests	130-113-333

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 DB105

Isotype mouse IgG1κ

Isotype control Mouse IgG1 – isotype control antibodies

Alternative names of antigen CSPG9, EMCR III, HCELL, HUTCH-I, CD44s, H-CAM, Pgp-1

Entrez Gene ID 960
Molecular mass of antigen [kDa] 79

Cross-reactivity rhesus monkey (*Macaca mulatta*)

Distribution of antigen bone marrow, cancer stem cells, CNS cells, endothelial cells, epithelial cells, kidney,

1

leukocytes, lymphocytes, mesenchymal stem cells, myeloid cells, plasma cells, ES and

iPS cells, red blood cells, skeletal muscle, skin, T cells

Product formatReagents 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.

Clone DB105 recognizes the CD44 antigen. CD44 is a marker for many types of cancer stem cells (CSCs), including breast CSCs that possess higher tumorigenicity and metastatic potential, colorectal, pancreatic, and prostate CSCs. In addition, expression was observed in several cancers as well as on carcinoma cell lines. Here, CD44 plays a role in cancer cell migration and matrix adhesion in response to a cellular microenvironment, thus enhancing cellular aggregation and tumor cell growth. CD44 is also expressed on mesodermal cells, such as hematopoietic, fibroblastic, and glial 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 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.

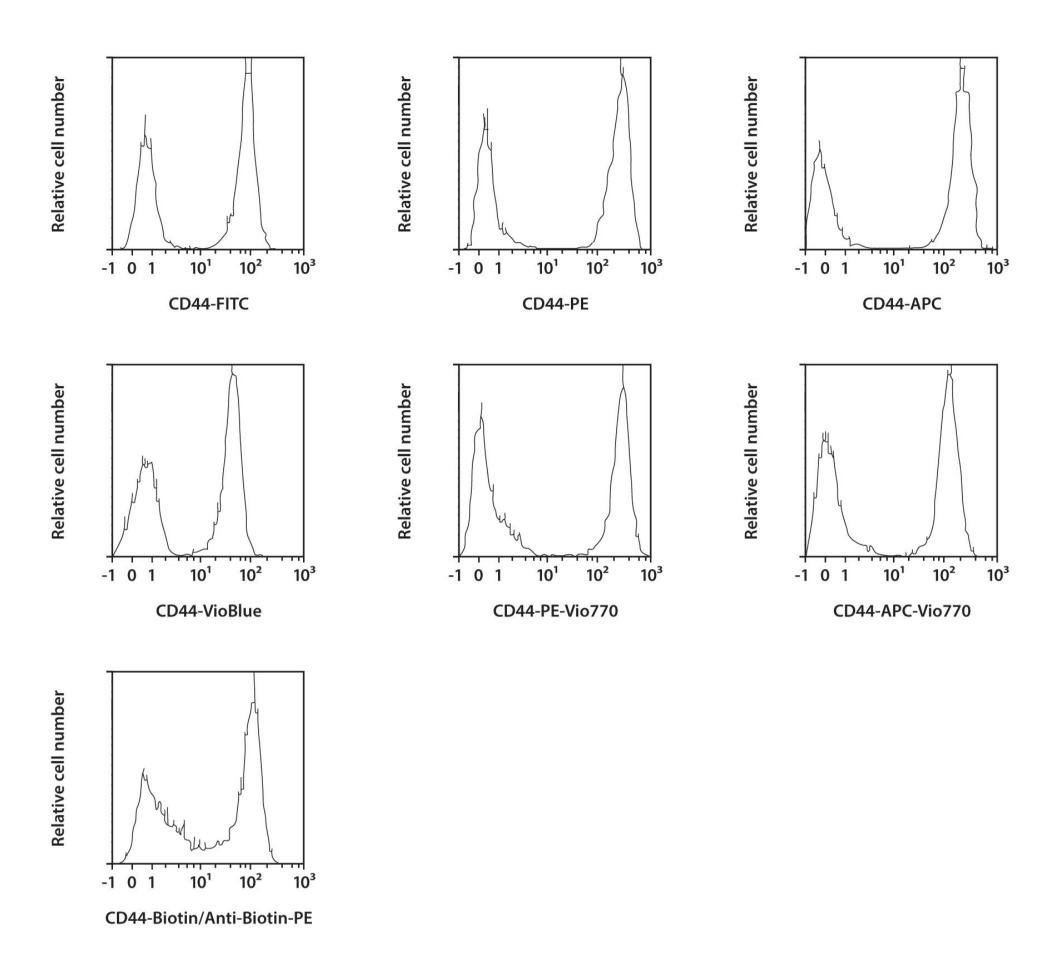
Protocol for cell surface staining

- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:50 for up to 10^6 cells/100 μ L.
- Volumes given below are for up to 10[°] nucleated cells. When working with fewer than 10[°] 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 $300\times g$ for 10 minutes. Aspirate supernatant completely.
- 3. Resuspend up to 10^6 nucleated cells per 98 μL of buffer.
- 4. Add 2 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 $300 \times g$ 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

A mixture of cells from U937 (CD44₊) and 1881 (CD44₋) cell lines was stained with CD44 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. 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 tandem conjugates.



Warranty

The products sold hereunder are warranted only to be free from defects in workmanship and material at the time of delivery to the customer. Miltenyi Biotec GmbH makes no warranty or representation, either expressed or implied, with respect to the fitness of a product for a particular purpose. There are no warranties, expressed or implied, which extend beyond the technical specifications of the products. Miltenyi Biotec GmbH's liability is limited to either replacement of the products or refund of the purchase price. Miltenyi Biotec GmbH is not liable for any property damage, personal injury or economic loss caused by the product.

Miltenyi Biotec GmbH | Friedrich-Ebert-Straße 68 | 51429 Bergisch Gladbach | Germany | Phone +49 2204 8306-0 | Fax +49 2204 85197 | macs@miltenyibiotec.de | www.miltenyibiotec.com Miltenyi Biotec provides products and services worldwide. Visit www.miltenyibiotec.com/local to find your nearest Miltenyi Biotec contact.

Unless otherwise specifically indicated, Miltenyi Biotec products and services are for research use only and not for therapeutic or diagnostic use. autoMACS, MACS, MACSQuant, Vio, VioBlue, VioBright, and VioGreen are either trademarks or registered trademarks of Miltenyi Biotec GmbH. Copyright © 2016 Miltenyi Biotec GmbH. All rights reserved.

3