CD163 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.
CD163-FITC	for 30 tests	130-099-969
CD163-FITC	for 100 tests	130-097-626
CD163-PE	for 100 tests	130-097-628
CD163-APC	for 30 tests	130-100-612
CD163-APC	for 100 tests	130-097-630
CD163-VioBlue	for 30 tests	130-099-973
CD163-VioBlue	for 100 tests	130-097-624
CD163-PE-Vio770	for 30 tests	130-101-546
CD163-PE-Vio770	for 100 tests	130-101-484
CD163-Biotin	for 100 tests	130-097-622

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	CD163
Clone	GHI/61.1
Isotype	mouse IgG1κ
Isotype control	Mouse IgG1 – isotype control antibodies
Alternative names of antigen	GHI/61, M130, MM130, CD163
Molecular mass of antigen [kDa]	121
Cross-reactivity	cynomolgus monkey (*Macaca fascicularis*)
Distribution of antigen	bone marrow, CNS cells, liver, macrophages, monocytes, spleen, thymocytes
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 monoclonal antibody GHI/61.1 reacts with human CD163, a single-chain transmembrane protein also known as hemoglobin scavenger receptor or M130. It is expressed by mature tissue macrophages and peripheral blood monocytes. The expression of CD163 is up-regulated *in vitro* and *in vivo* by

anti-inflammatory mediators such as IL-10 and (gluco)corticosteroid and is shed to blood upon inflammatory activation of macrophages. CD163 functions as a high affinity scavenger receptor for the complex of haemoglobin and haptoglobin. Depending on the ligand, crosslinking of M130 initiates signal transduction leading to the production of proinflammatory cytokines Il-1ß, IL-6, and GM-CSF or the antiinflammatory cytokine IL-10.

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.

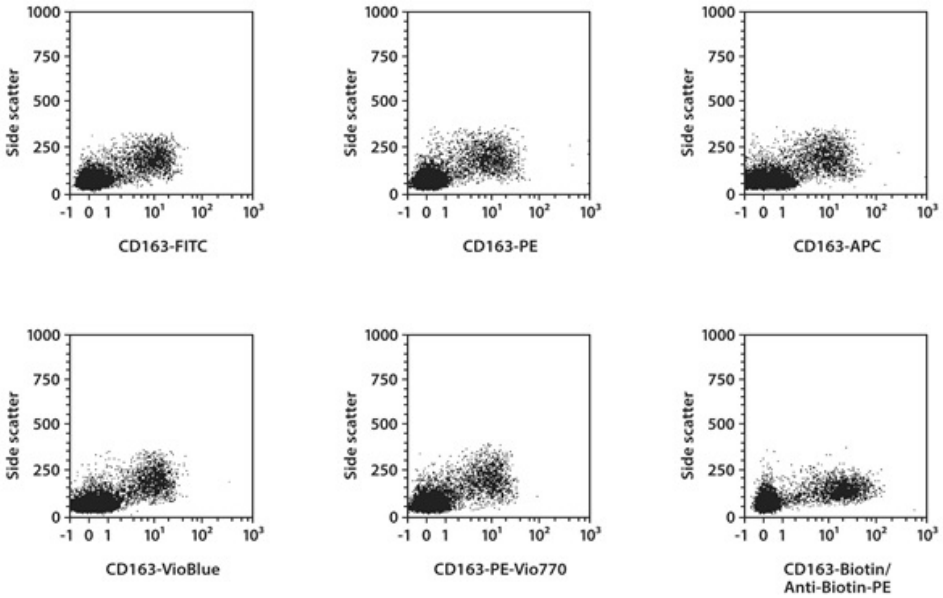
Protocol for cell surface staining

- 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 CD163 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.



References

1. **Kristiansen, M. *et al.*** (2001) Identification of the haemoglobin scavenger receptor. Nature 409(6817): 198–201.
2. **Law, S. K. *et al.*** (1993) A new macrophage differentiation antigen which is a member of the scavenger receptor superfamily. Eur. J. Immunol. 23(9): 2320–2325.
3. **Ritter, M. *et al.*** (2001) Interaction of CD163 with the regulatory subunit of casein kinase II (CKII) and dependence of CD163 signaling on CKII and protein kinase C. Eur. J. Immunol. 31(4): 999–1009.
4. **Pulford, K. *et al.*** (1992) A monocyte/macrophage antigen recognized by the four antibodies GHI/61, Ber-MAC3, Ki-M8 and SM4. Immunology 75(4): 588–595.
5. Burdo, T. H. *et al.* (2011) Soluble CD163 made by monocyte/macrophages is a novel marker of HIV activity in early and chronic infection prior to and after anti-retroviral therapy. J. Infect. Dis. 204: 154–163.

Warranty

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