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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.

1. Description

This product is for research use only.

Components Monoclonal CD11b antibodies, human and mouse conjugated to:

Conjugate	Order no. 1 mL (100 tests)	Order no. 300 µL (30 tests)
FITC	130-081-201	130-098-085
PE	130-091-240	130-098-087
APC	130-091-241	130-098-088
VioBlue®	130-097-336	130-098-086
VioGreen™	130-097-299	130-098-090
PE-Vio770™	130-099-704	130-099-708
APC-Vio770™	130-096-834	130-098-089
PerCP-Vio700™	130-097-585	–
Biotin	130-098-581	130-098-582

Clone M1/70.15.11.5 (isotype: rat IgG2b).

Capacity 1 mL: 100 tests or up to 10⁹ total cells
300 µL: 30 tests or up to 3×10⁸ total cells.

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

Storage Store protected from light at 2–8 °C. Do not freeze. The expiration date is indicated on the vial label.

Cross-reactivity: The CD11b antibody has been reported to react with

- rhesus monkey (*Macaca mulatta*) cells
- cynomolgus monkey (*Macaca fascicularis*) cells

1.1 Background information

- Antigen: CD11b
- Synonym: integrin αM; Mac-1
- Expression patterns: CD11b (Mac-1 α; integrin αM chain) is part of the CD11b/CD18 heterodimer (Mac-1 α, Mβ2 integrin), also known as the C3 complement receptor. It functions as a receptor for complement (C3bi), fibrinogen, or clotting factor X. In humans, CD11b is strongly expressed on myeloid cells and weakly expressed on NK cells and some activated lymphocytes as well as on microglia in the brain. In mice, the CD11b antigen is expressed on monocytes/macrophages and microglia. To a lower extent it is expressed on granulocytes, NK cells, CD5⁺ B-1 cells, and subsets of dendritic cells. The monoclonal M1/70.15.11.5 antibody recognizes the human, mouse, and non-human primate CD11b antigen.

1.2 Applications

- Identification and enumeration of CD11b⁺ cells by flow cytometry.

1.3 Recommended antibody dilution

The recommended antibody dilution for all CD11b conjugates is **1:11 for up to 10⁷ cells/100 µL** of buffer for labeling of cells and subsequent analysis by flow cytometry.

Cells should be stained prior to fixation, if formaldehyde is used as a fixative.

1.4 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) Conjugated anti-biotin antibodies, e.g., Anti-Biotin-PE (# 130-090-756) as secondary antibody reagent in combination with CD11b-Biotin.
- (Optional) For antibodies for additional staining or for isotype control, refer to www.miltenyibiotec.com/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.

2. General protocol for immunofluorescent staining

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).

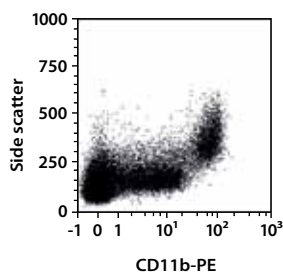
1. Determine cell number.
2. Centrifuge cell suspension at $300 \times g$ for 10 minutes. Aspirate supernatant completely.
3. Resuspend up to 10^7 nucleated cells per 100 μL of buffer.
4. Add 10 μL of the CD11b antibody.
5. Mix well and incubate for 10 minutes in the dark in the refrigerator ($2-8^\circ\text{C}$).

▲ **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 CD11b-Biotin was used, resuspend the cell pellet in 100 μL of buffer, add 10 μL of 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.

3. Example of immunofluorescent staining with CD11b antibodies

Human peripheral blood mononuclear cells (PBMCs) were stained with CD11b antibodies conjugated to PE and analyzed by flow cytometry using the MACSQuant® Analyzer. Cell debris and dead cells were excluded from the analysis based on scatter signals and propidium iodide fluorescence.



For more examples please refer to the respective product page at www.miltenyibiotec.com/antibodies.

Refer to www.miltenyibiotec.com for all data sheets and protocols.

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