Miltenyi Biotec

Anti-Biotin antibodies

Contents

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 Anti-Biotin antibodies, conjugated to:

Conjugate	Order no. 1 mL (100 tests)	Order no. 300 µL (30 tests)
FITC	130-090-857	130-098-796
VioBright™ FITC	130-104-514	130-104-563
PE	130-090-756	–
APC	130-090-856	–
VioBlue®	130-094-669	130-098-800
VioGreen™	130-097-022	130-099-129
PerCP	130-094-974	130-098-799
PE-Vio® 615	130-107-140	130-107-195
PE-Vio770	130-096-632	130-099-237
APC-Vio770	130-096-630	130-099-122
PerCP-Vio700	130-097-587	–

Clone Bio3-18E7 (isotype: mouse IgG1).

Capacity 1 mL: 100 tests or up to 10^9 total cells

300 µL: 30 tests or up to 3×10^8 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.

1.1 Background information

- Antigen: Biotin
- Expression patterns: Anti-Biotin fluorochromes are ideal for fluorescent staining of cells separated with Anti-Biotin MicroBeads, Streptavidin MicroBeads, or with the Anti-Biotin MultiSort Kit. They can also be used to stain cells labeled with cocktails of biotinylated antibodies, for example for quality control when using MACS® Cell Isolation Kits for the isolation of untouched cells. The Anti-Biotin antibody does not bind to free biotin which is often present in cell culture media.

1.2 Applications

- Fluorescent staining of cells labeled with a biotinylated primary antibody.
- Fluorescent staining of cells separated with Anti-Biotin MicroBeads (# 130-090-485) or Streptavidin MicroBeads (# 130-048-101).
- Quality control of separations using MACS Technology.

1.3 Recommended antibody dilution

The recommended antibody dilution for all Anti-Biotin conjugates is **1:11 for up to 10^7 cells/100 µL** of buffer for labeling of cells and subsequent analysis by flow cytometry. For Anti-Biotin MicroBead-labeled cells use the same dilution. Staining intensity depends on biotinylation grade of the primary antibody.

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

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 Ca^{2+} or Mg^{2+} are not recommended for use.

- (Optional) Tandem Signal Enhancer, human (# 130-099-888) to reduce non-specific binding of tandem dye–conjugated antibodies to human cells, especially to monocytes.
- (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.

140-002-163.07

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2. General protocol for immunofluorescent staining

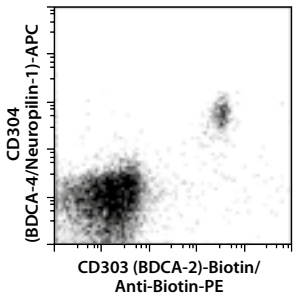
Volumes given below are for **up to 10^7** nucleated cells. When working with fewer than 10^7 cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes accordingly (e.g. for 2×10^7 nucleated cells, use twice the volume of all indicated reagent volumes and total volumes).

1. Determine cell number.
2. Centrifuge cell suspension at 300×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 Anti-Biotin 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. Resuspend cell pellet in a suitable amount of buffer for analysis by flow cytometry or fluorescence microscopy.

3. Example of immunofluorescent staining with Anti-Biotin antibodies

Human peripheral blood mononuclear cells (PBMCs) were labeled with CD303 (BDCA-2)-Biotin and fluorescently stained with Anti-Biotin antibodies conjugated to PE. Cells were counterstained with CD304 (BDCA-4/Neuropilin-1)-APC and analyzed by flow cytometry using the MACSQuant® Analyzer.



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.

Warranty