# CD1c (BDCA-1) 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. |
|---|---|---|
| CD1c (BDCA-1)-FITC[1] | for 30 tests | 130-098-009 |
| CD1c (BDCA-1)-FITC[1] | for 100 tests | 130-090-507 |
| CD1c (BDCA-1)-VioBright FITC | for 30 tests | 130-104-894 |
| CD1c (BDCA-1)-VioBright FITC | for 100 tests | 130-104-848 |
| CD1c (BDCA-1)-PE | for 30 tests | 130-098-007 |
| CD1c (BDCA-1)-PE | for 100 tests | 130-090-508 |
| CD1c (BDCA-1)-APC | for 30 tests | 130-098-005 |
| CD1c (BDCA-1)-APC | for 100 tests | 130-090-903 |
| CD1c (BDCA-1)-PE-Vio770 | for 30 tests | 130-101-582 |
| CD1c (BDCA-1)-PE-Vio770 | for 100 tests | 130-101-581 |
| CD1c (BDCA-1)-Biotin[1] | for 30 tests | 130-098-011 |
| CD1c (BDCA-1)-Biotin[1] | for 100 tests | 130-090-692 |
| CD1c (BDCA-1) pure | 100 µg in 1 mL | 130-090-695 |

[1]Not recommended for cells that are labeled with MACS MicroBeads using the same antigen.

## 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

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|---|---|
| **Antigen** | CD1c (BDCA-1) |
| **Clone** | AD5-8E7 |
| **Isotype** | mouse IgG2a |
| **Isotype control** | Mouse IgG2a – isotype control antibodies |
| **Alternative names of antigen** | BDCA-1, CD1, R7, R7, M241, T6, CD1a |
| **Molecular mass of antigen [kDa]** | 36 |
| **Cross-reactivity** | rhesus monkey (*Macaca mulatta*), cynomolgus monkey (*Macaca fascicularis*) |
| **Distribution of antigen** | B cells, dendritic cells, Langerhans cells, lymphocytes, macrophages, monocytes, myeloid leukemia cells, T cells, thymocytes |
| **Product format** | Antibodies are supplied in buffer containing stabilizer and 0.05% sodium azide. |

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|---|---|
| **Fixation** | Cells should be stained prior to fixation, if formaldehyde is used as a fixative. |
| **Storage** | Store protected from light at 2–8 °C. Do not freeze. |

The clone AD5-8E7 recognizes human CD1c, also known as BDCA-1. CD1c is expressed on a major subpopulation of human myeloid dendritic cells (about 0.3% of blood leukocytes). Blood CD1c (BDCA-1)$^{+}$ myeloid dendritic cells are CD11c$^{high}$, CD123$^{low}$, CD4$^{+}$, Lin$^{–}$, CD45RO$^{+}$, CD2$^{+}$, CD16$^{–}$, CD141 (BDCA-3)$^{low}$, CD303 (BDCA-2)$^{–}$, and CD304 (BDCA-4/Neuropilin-1)$^{–}$. They express myeloid markers (CD13, CD33) as well as Fc receptors (CD32, CD64, FcεRI) and are of monocytoid appearance. A minor proportion of CD1c (BDCA-1)$^{+}$ myeloid dendritic cells expresses CD14 and CD11b. In blood, CD1c (BDCA-1) is also expressed on a subpopulation of CD19$^{+}$ small resting B lymphocytes. CD1c (BDCA-1) expression has also been detected on cortical thymocytes, on Langerhans cells, and on CD1a$^{+}$ dendritic cells generated *ex vivo* from monocytes or hematopoietic precursor cells. CD1c (BDCA-1)$^{+}$ myeloid dendritic cells have been designated as type-1 myeloid dendritic cells (MDC1s).
The CD1c antigen is a member of the CD1 family of proteins that are structurally related to MHC class I proteins and mediate the presentation of non-peptide antigens to 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, 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.

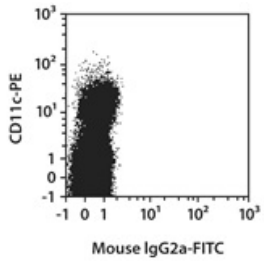
## Protocol for cell surface staining

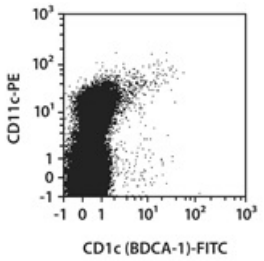
- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:11 for up to $10^7$ cells/100 µL of buffer.
- Volumes given below are for up to $10^7$ nucleated cells. When working with fewer than $10^7$ cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes accordingly (e.g. for $2\times10^7$ nucleated cells, use twice the volume of all indicated reagent volumes and total volumes).

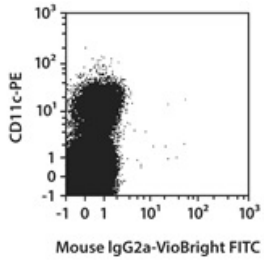
1. Determine cell number.
2. Centrifuge cell suspension at 300×g for 10 minutes. Aspirate supernatant completely.
3. Resuspend up to $10^7$ nucleated cells per 100 µL of buffer.
4. Add 10 µL of the antibody.
5. Mix well and incubate for 10 minutes in the dark in the refrigerator (2−8 °C).
   Note: Higher 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.

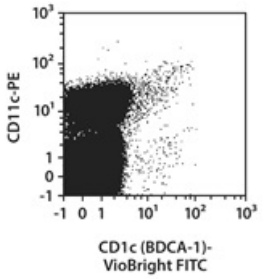
## Examples of immunofluorescent staining

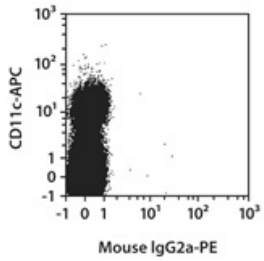
Human peripheral blood mononuclear cells (PBMCs) were stained with CD1c (BDCA-1) antibodies or with the corresponding isotype control antibodies (left image), as well as with CD19 and CD11c antibodies. The Tandem Signal Enhancer has been used to increase binding specificity of tandem-dye–conjugated antibodies. For all other conjugates the FcR Blocking Reagent has been used to avoid Fc receptor–mediated antibody labeling. Apart from type-1 myeloid dendritic cells (MDC1s), CD1c (BDCA-1) is also expressed on a subset of B cells. For this reason, B cells are excluded from the analysis according to expression of CD19 and further analysis is restricted to CD19– cells. Cell debris and dead cells were excluded from the analysis based on scatter signals and propidium iodide or 4',6-diamidino-2-phenylindole (DAPI) fluorescence, as in the case of tandem conjugates.

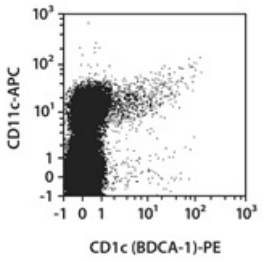

CD11c-PE
Mouse IgG2a-FITC

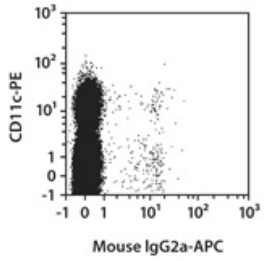

CD11c-PE
CD1c (BDCA-1)-FITC

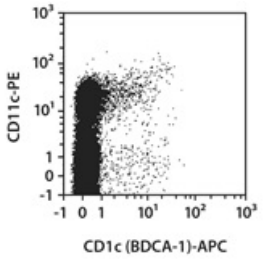

CD11c-PE
Mouse IgG2a-VioBright FITC

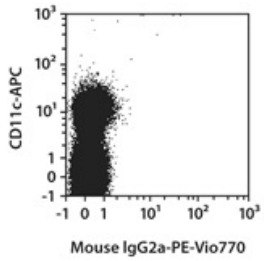

CD11c-PE
CD1c (BDCA-1)-
VioBright FITC

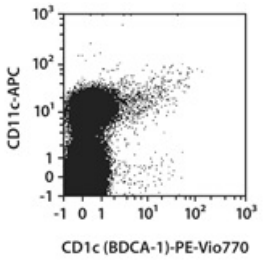

CD11c-APC
Mouse IgG2a-PE

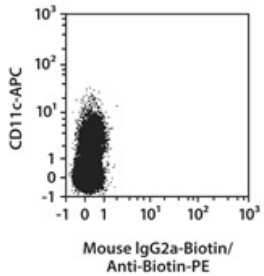

CD11c-APC
CD1c (BDCA-1)-PE

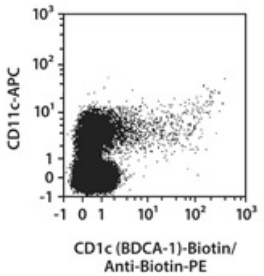

CD11c-PE
Mouse IgG2a-APC


CD11c-PE
CD1c (BDCA-1)-APC

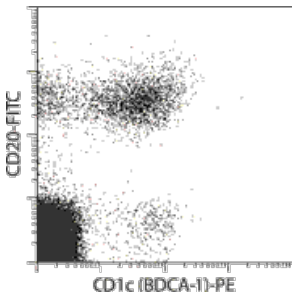

CD11c-APC
Mouse IgG2a-PE-Vio770


CD11c-APC
CD1c (BDCA-1)-PE-Vio770

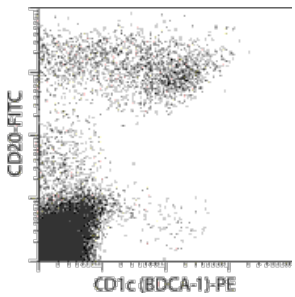

CD11c-APC
Mouse IgG2a-Biotin/
Anti-Biotin-PE


CD11c-APC
CD1c (BDCA-1)-Biotin/
Anti-Biotin-PE

PBMCs from rhesus monkey were stained with CD1c (BDCA-1)-PE and CD20-FITC and analyzed by flow cytometry.




Cynomolgus monkey PBMCs were stained with CD1c (BDCA-1)-PE and CD20-FITC and analyzed by flow cytometry.

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