

# CD1a antibodies, human

**For research use only**

One test corresponds to labeling of up to  $10^7$  cells in a total volume of 100  $\mu$ L.

Product	Content	Order no.
CD1a-FITC	for 100 tests	130-097-903
CD1a-PE	for 100 tests	130-097-868
CD1a-APC	for 100 tests	130-097-875
CD1a-VioBlue	for 100 tests	130-097-905
CD1a-PE-Vio770	for 30 tests	130-105-579
CD1a-PE-Vio770	for 100 tests	130-105-527
CD1a-APC-Vio770	for 30 tests	130-100-225
CD1a-APC-Vio770	for 100 tests	130-100-224
CD1a-Biotin	for 100 tests	130-097-908

## 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

<b>Antigen</b>	CD1a
<b>Clone</b>	HI149
<b>Isotype</b>	mouse IgG1 $\kappa$
<b>Isotype control</b>	Mouse IgG1 – isotype control antibodies
<b>Alternative names of antigen</b>	HTA1, CD1, FCB6, HTA1, R4, T6, Leu-6, CD1a
<b>Molecular mass of antigen [kDa]</b>	35
<b>Distribution of antigen</b>	B cells, dendritic cells, Langerhans cells, lymphocytes, macrophages, monocytes, thymocytes
<b>Product format</b>	Antibodies are supplied in buffer containing stabilizer and 0.05% sodium azide.
<b>Fixation</b>	The antibody is suited for staining of formaldehyde-fixed cells.
<b>Storage</b>	Store protected from light at 2–8 °C. Do not freeze.

Clone HI149 reacts with human CD1a, a 40kDa type I membrane glycoprotein also known as T6 which is a member of the immunoglobulin superfamily. It shares functional and structural similarities with MHC I class molecules and is associated with  $\beta$ 2-microglobulin. CD1a plays a role in antigen presentation. It is expressed on Langerhans cells, dendritic cells, and cortical thymocytes.

## Reagent requirements

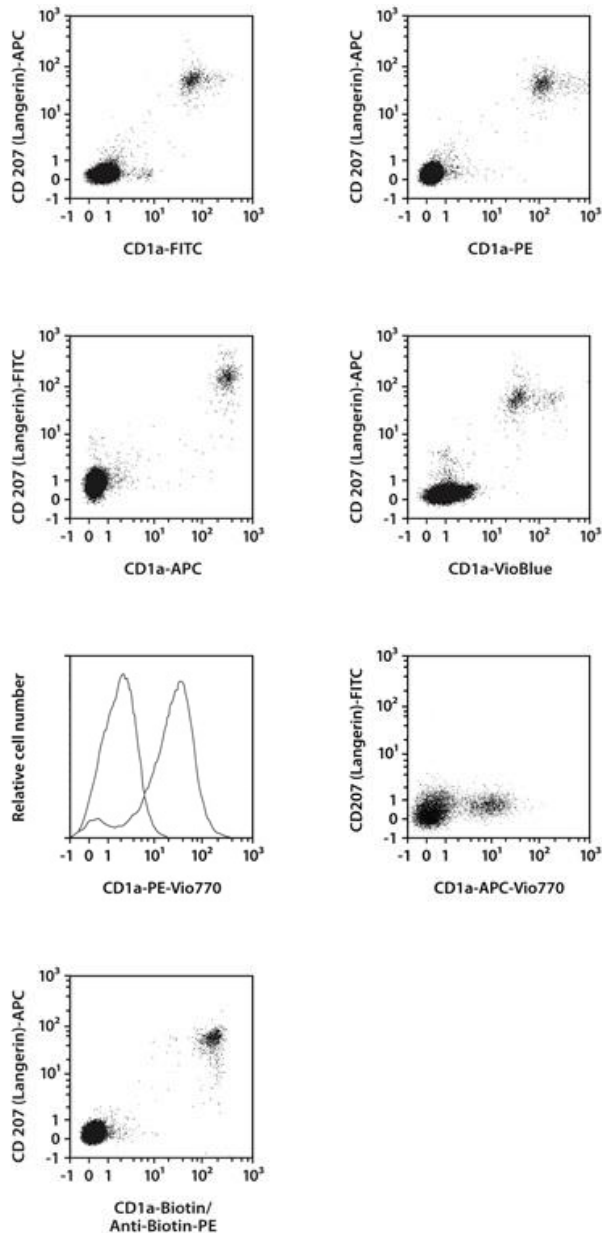
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS<sup>®</sup> BSA Stock Solution (# 130-091-376) 1:20 with autoMACS<sup>®</sup> Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).  
Note: EDTA can be replaced by other supplements such as anticoagulant citrate dextrose formula-A (ACD-A) or citrate phosphate dextrose (CPD). Buffers or media containing Ca<sup>2+</sup> or Mg<sup>2+</sup> 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<sup>7</sup> cells/100 µL of buffer.
  - Volumes given below are for up to 10<sup>7</sup> nucleated cells. When working with fewer than 10<sup>7</sup> 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<sup>7</sup> 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<sup>7</sup> 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 epidermal cells were stained with CD1a antibodies as well as with CD207 (Langerin) antibodies and analyzed by flow cytometry using the MACSQuant<sup>®</sup> Analyzer. 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.



## References

1. **Blumberg, R. S. et al.** (1995) Structure and function of the CD1 family of MHC-like cell surface proteins. *Immunol. Rev.* 147: 5–29.
2. **Hanau, D. et al.** (1990) Possible mechanism of action of CD1a antigens. *J. Invest. Dermatol.* 95(5): 503–505.
3. **Calabi, F. et al.** (1991) The CD1 system. *Tissue Antigens* 37(1): 1–9.

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