CD159a (NKG2A) 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.
CD159a (NKG2A)-FITC	for 30 tests	130-098-817
CD159a (NKG2A)-FITC	for 100 tests	130-098-818
CD159a (NKG2A)-VioBright FITC	for 30 tests	130-105-693
CD159a (NKG2A)-VioBright FITC	for 100 tests	130-105-646
CD159a (NKG2A)-PE	for 30 tests	130-098-813
CD159a (NKG2A)-PE	for 100 tests	130-098-814
CD159a (NKG2A)-APC	for 30 tests	130-098-809
CD159a (NKG2A)-APC	for 100 tests	130-098-812
CD159a (NKG2A)-PE-Vio770	for 30 tests	130-105-694
CD159a (NKG2A)-PE-Vio770	for 100 tests	130-105-647
CD159a (NKG2A)-Biotin	for 30 tests	130-098-822
CD159a (NKG2A)-Biotin	for 100 tests	130-098-819

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	CD159a (NKG2A)
Clone	REA110
Isotype	recombinant human IgG1
Isotype control	REA Control (S) antibodies
Alternative names of antigen	KLRC1, CD159a, NKG2, NKG2A
Molecular mass of antigen [kDa]	26
Cross-reactivity	chimpanzee (*Pan troglodytes*), olive baboon (*Papio anubis*), rhesus monkey (*Macaca mulatta*), cynomolgus monkey (*Macaca fascicularis*)
Distribution of antigen	NK cells, T cells
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.

Clone REA110 recognizes CD159a, an inhibitory natural killer (NK) cell receptor. CD159a forms heterodimer with CD94 and contains a C-type lectin ectodomain. Inhibitory signal is transmitted by the heterodimer via immunoreceptor tyrosine-based inhibitory motifs (ITIMs) and upon ligand engagement, ITIMs are phosphorylated and transmit signal through docking of various tyrosine phosphatases. CD159a/CD94 binds non-classical MHC class I protein, HLA-E. Expression of CD159a is found mainly on NK cells and on subsets of $CD8^+$ cells.
Additional information: Clone REA110 displays negligible binding to Fc receptors.

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) 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.

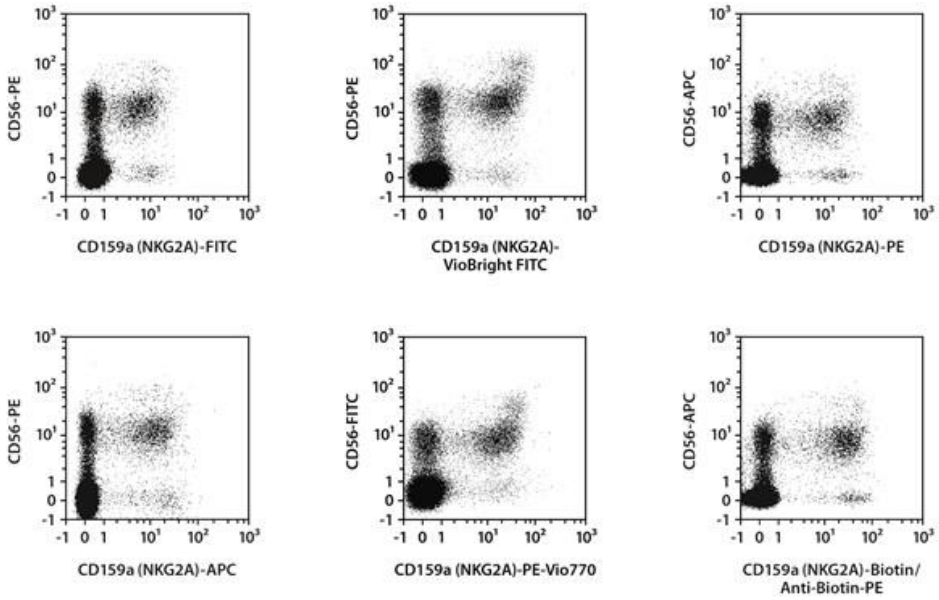
Protocol for cell surface staining

- 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 CD159a (NKG2A) antibodies as well as with CD56 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. **Boyington, J. C. *et al.*** (1999) Structure of CD94 reveals a novel C-type lectin fold: implications for the NK cell-associated CD94/NKG2 receptors. Immunity 10(1): 75–82.
2. **Sullivan, L. C. *et al.*** (2007) The heterodimeric assembly of the CD94-NKG2 receptor family and implications for human leukocyte antigen-E recognition. Immunity 27(6): 900–911.
3. **Borrego, F., *et al.*** (2006) The CD94/NKG2 family of receptors: from molecules and cells to clinical relevance. Immunol. Res. 35(3): 263–278.

Warranty

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