

CD158e/k (KIR3DL1/DL2) antibodies, human

For research use only

One test corresponds to labeling of up to 10^6 cells in a total volume of 100 μ L

Product	Content	Order no.
CD158e/k (KIR3DL1/DL2)-Biotin	for 100 tests	130-116-176
CD158e/k (KIR3DL1/DL2)-FITC	for 30 tests	130-116-280
CD158e/k (KIR3DL1/DL2)-FITC	for 100 tests	130-116-177
CD158e/k (KIR3DL1/DL2)-PE	for 30 tests	130-116-281
CD158e/k (KIR3DL1/DL2)-PE	for 100 tests	130-116-178
CD158e/k (KIR3DL1/DL2)-APC	for 30 tests	130-116-282
CD158e/k (KIR3DL1/DL2)-APC	for 100 tests	130-116-179
CD158e/k (KIR3DL1/DL2)-PE-Vio615	for 30 tests	130-116-286
CD158e/k (KIR3DL1/DL2)-PE-Vio615	for 100 tests	130-116-183
CD158e/k (KIR3DL1/DL2)-PE-Vio770	for 30 tests	130-116-283
CD158e/k (KIR3DL1/DL2)-PE-Vio770	for 100 tests	130-116-180
CD158e/k (KIR3DL1/DL2)-APC-Vio770	for 100 tests	130-116-181
CD158e/k (KIR3DL1/DL2)-Biotin	for 30 tests	130-116-279

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	CD158e/k (KIR3DL1/DL2)
Clone	REA970
Isotype	recombinant human IgG1
Isotype control	REA Control (S) antibodies
Alternative names of antigen	KIR3DL1, KIR3DL2
Entrez Gene ID	3811
Distribution of antigen	NK cells, T cells
Product format	Reagents are supplied in buffer containing stabilizer and 0.05% sodium azide.
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.

Clone REA970 recognizes the human CD158e (KIR3DL1) and CD158k (KIR3DL2) antigens, members of the killer immunoglobulin-like receptor (KIR) family which recognizes subsets of HLA alleles. The family of KIR contributes to the regulation of NK cell-mediated cytotoxicity. Expression is found mainly on CD56^{dim} CD16⁺ natural killer (NK) cells but also on a subset of CD8⁺ T cells. They are monomeric receptors possessing high allelic polymorphism with either two or three Ig-like extracellular domains. According to the length of their cytoplasmic tail, KIRs can be subdivided in inhibitory KIRs and activating KIRs.

Additional information: Clone REA970 displays negligible binding to Fc receptors.

Reagent requirements

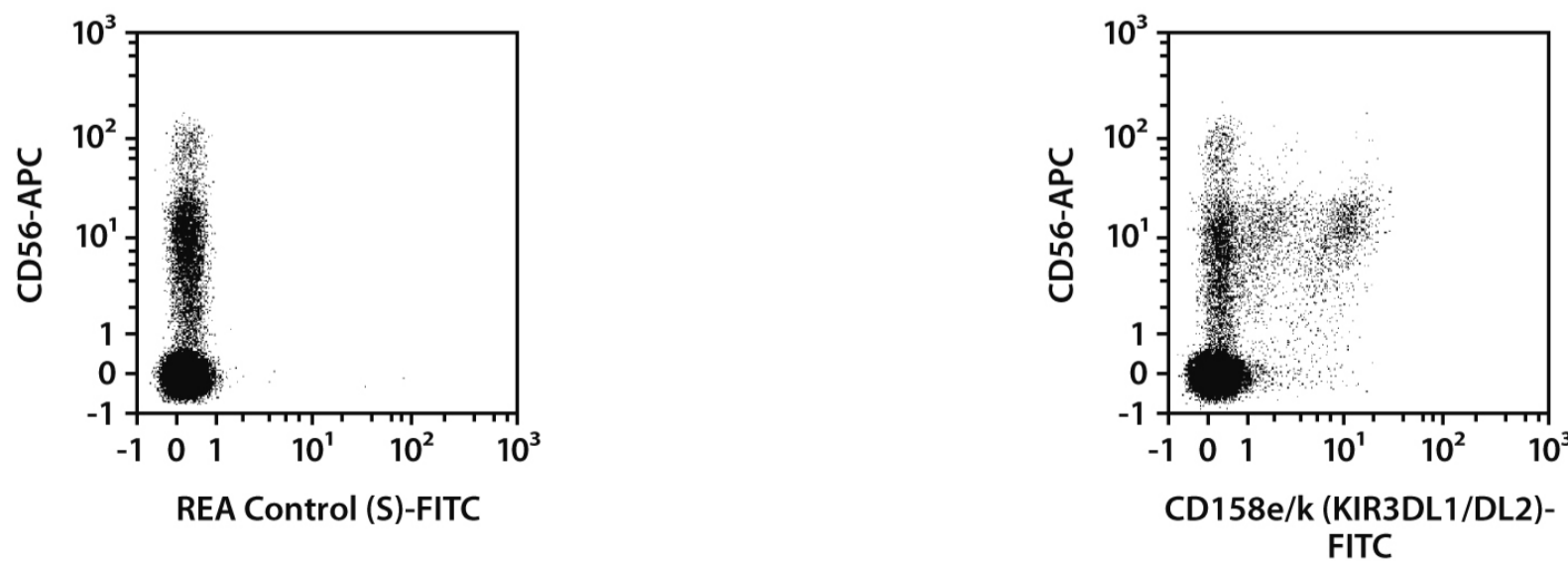
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS[®] BSA Stock Solution (# 130-091-376) 1:20 with autoMACS[®] Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).
Note: EDTA can be replaced by other supplements such as anticoagulant citrate dextrose formula-A (ACD-A) or citrate phosphate dextrose (CPD). Buffers or media containing Ca²⁺ or Mg²⁺ 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.

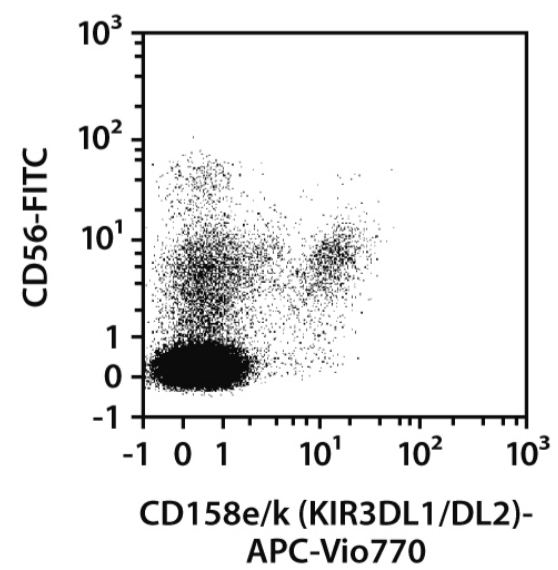
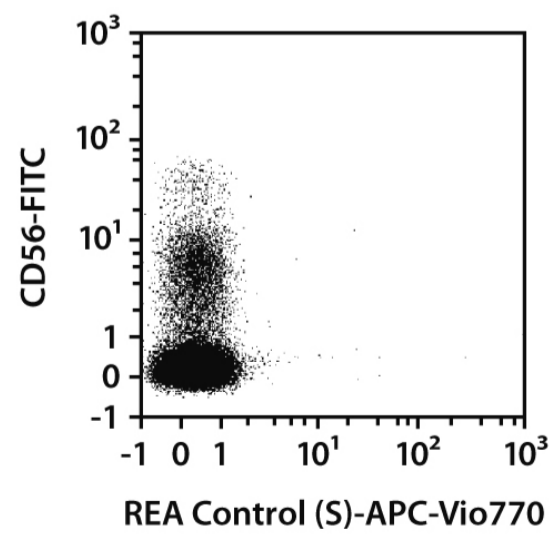
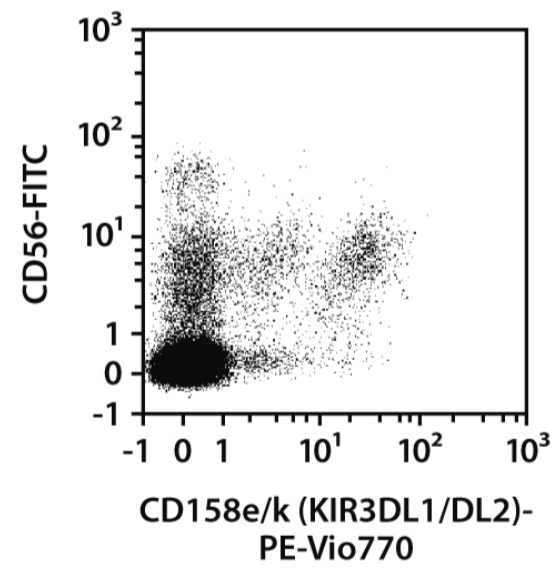
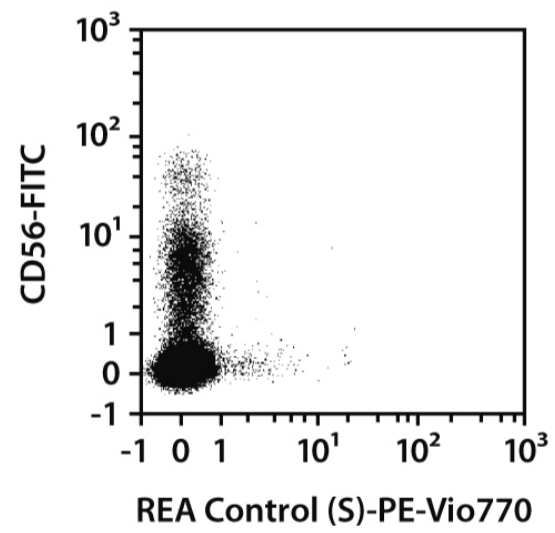
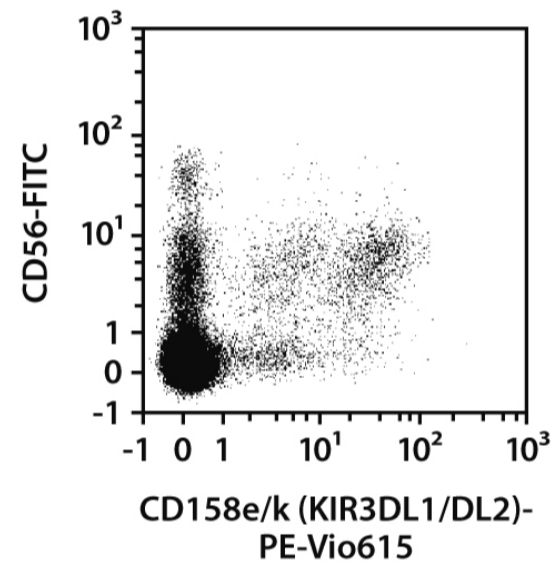
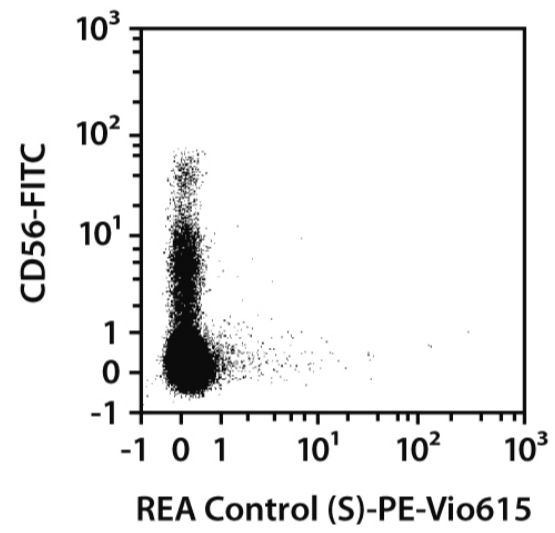
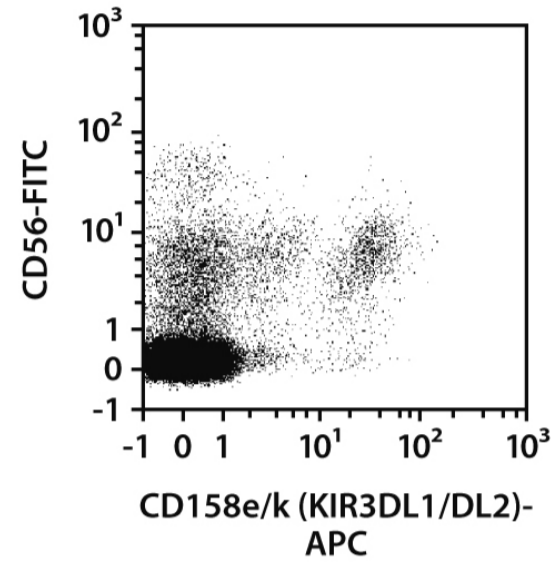
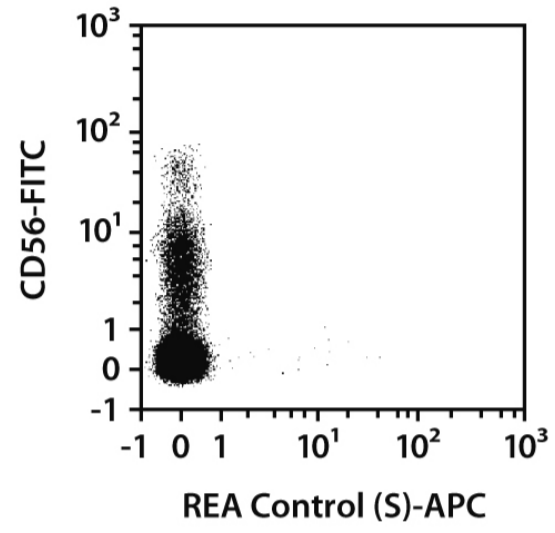
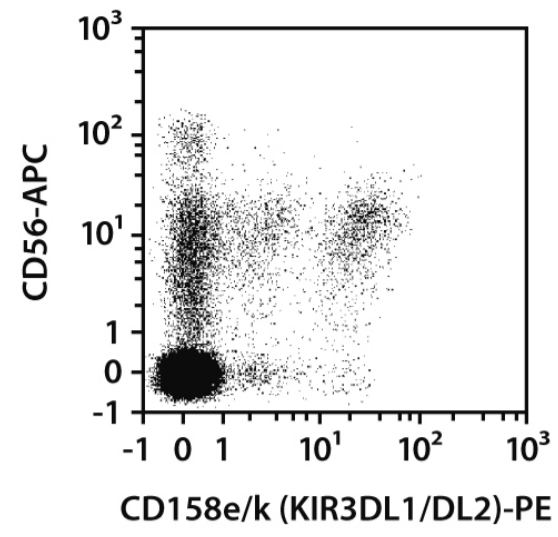
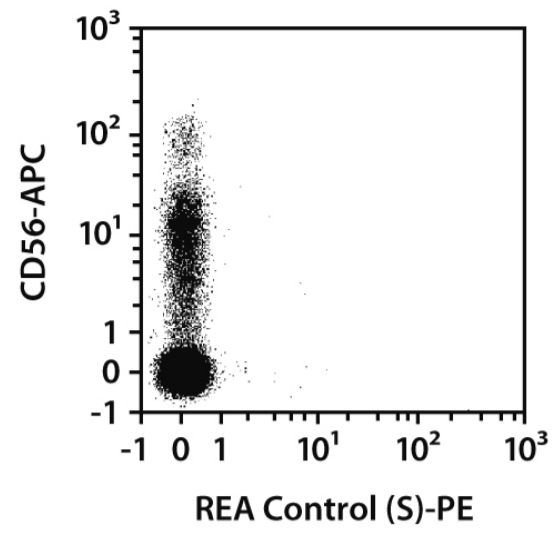
Protocol for cell surface staining

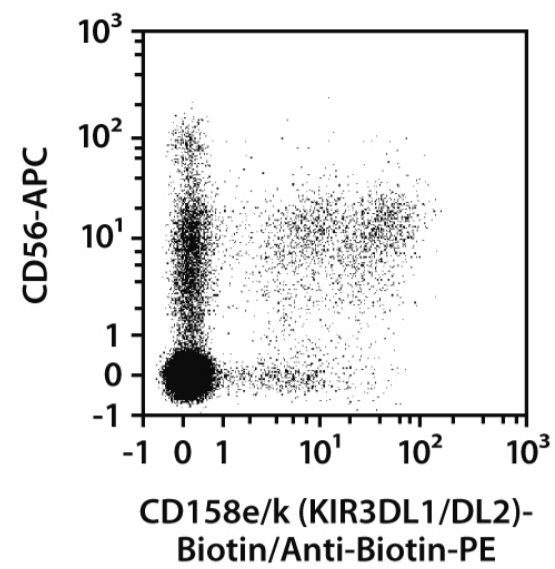
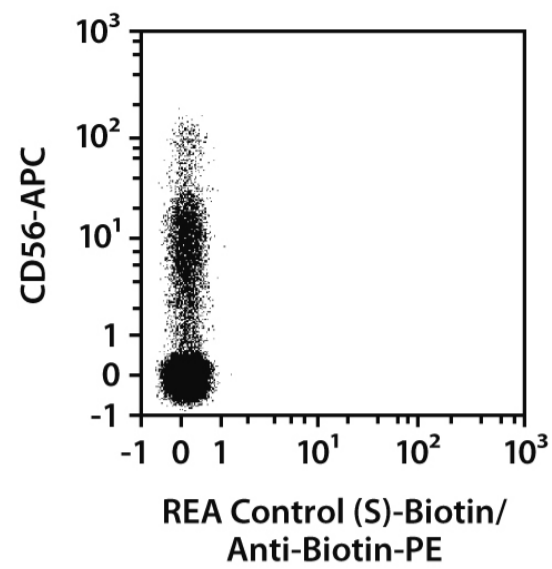
- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:50 for up to 10⁶ cells/100 µL.
 - Volumes given below are for up to 10⁶ nucleated cells. When working with fewer than 10⁶ cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes accordingly.
1. Determine cell number.
 2. Centrifuge cell suspension at 300×g for 10 minutes. Aspirate supernatant completely.
 3. Resuspend up to 10⁶ nucleated cells per 98 µL of buffer.
 4. Add 2 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 buffer and stain with fluorochrome-conjugated anti-biotin antibody according to the manufacturer's recommendations.
 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 cells (PBMCs) were stained with CD158e/k (KIR3DL1/DL2) antibodies or with the corresponding REA Control (S) antibodies (left images) as well as with CD56 antibodies. Flow cytometry was performed 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.







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Miltenyi Biotec GmbH | Friedrich-Ebert-Straße 68 | 51429 Bergisch Gladbach | Germany | Phone +49 2204 8306-0 | Fax +49 2204 85197 | macs@miltenyibiotec.de | www.miltenyibiotec.com Miltenyi Biotec provides products and services worldwide. Visit www.miltenyibiotec.com/local to find your nearest Miltenyi Biotec contact.

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