

# Anti-CLEC4D (CLECSF8) antibodies, human

## For research use only

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

Product	Content	Order no.
Anti-CLEC4D (CLECSF8)-PE-Vio770	for 100 tests	130-110-723
Anti-CLEC4D (CLECSF8)-PE	for 30 tests	130-110-858
Anti-CLEC4D (CLECSF8)-PE	for 100 tests	130-110-721
Anti-CLEC4D (CLECSF8)-APC	for 30 tests	130-110-859
Anti-CLEC4D (CLECSF8)-APC	for 100 tests	130-110-722
Anti-CLEC4D (CLECSF8)-PE-Vio770	for 30 tests	130-110-860
Anti-CLEC4D (CLECSF8)-APC-Vio770	for 30 tests	130-110-861
Anti-CLEC4D (CLECSF8)-APC-Vio770	for 100 tests	130-110-724
Anti-CLEC4D (CLECSF8)-VioBright 515	for 30 tests	130-111-663
Anti-CLEC4D (CLECSF8)-VioBright 515	for 100 tests	130-111-585
Anti-CLEC4D (CLECSF8)-Biotin	for 30 tests	130-110-857
Anti-CLEC4D (CLECSF8)-Biotin	for 100 tests	130-110-720

## 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>	CLEC4D (CLECSF8)
<b>Clone</b>	REA757
<b>Isotype</b>	recombinant human IgG1
<b>Isotype control</b>	REA Control (S) antibodies
<b>Alternative names of antigen</b>	CLECSF8, MCL, CLEC-6
<b>Entrez Gene ID</b>	<a href="#">338339</a>
<b>Molecular mass of antigen [kDa]</b>	25
<b>Distribution of antigen</b>	leukemia cells, bone marrow, spleen
<b>Product format</b>	Reagents are supplied in buffer containing stabilizer and 0.05% sodium azide.
<b>Fixation</b>	Cells should be stained prior to fixation, if formaldehyde is used as a fixative.
<b>Storage</b>	Store protected from light at 2-8 °C. Do not freeze.

Clone REA757 recognizes the human CLEC4D antigen, a cell-surface receptor that belongs to the human calcium-dependent lectin family (C-type lectin), also known as CLECSF8, CLEC-6, or MCL. CLEC4D is expressed in a low level in peripheral blood leukocytes, in bone marrow, and spleen. It is mainly found in monocytes and macrophage and seems to be up-regulated by IL-6,

IL-10, TNF- $\alpha$ , and IFN- $\gamma$ . Additional information: Clone REA757 displays negligible binding to Fc receptors.

## Reagent requirements

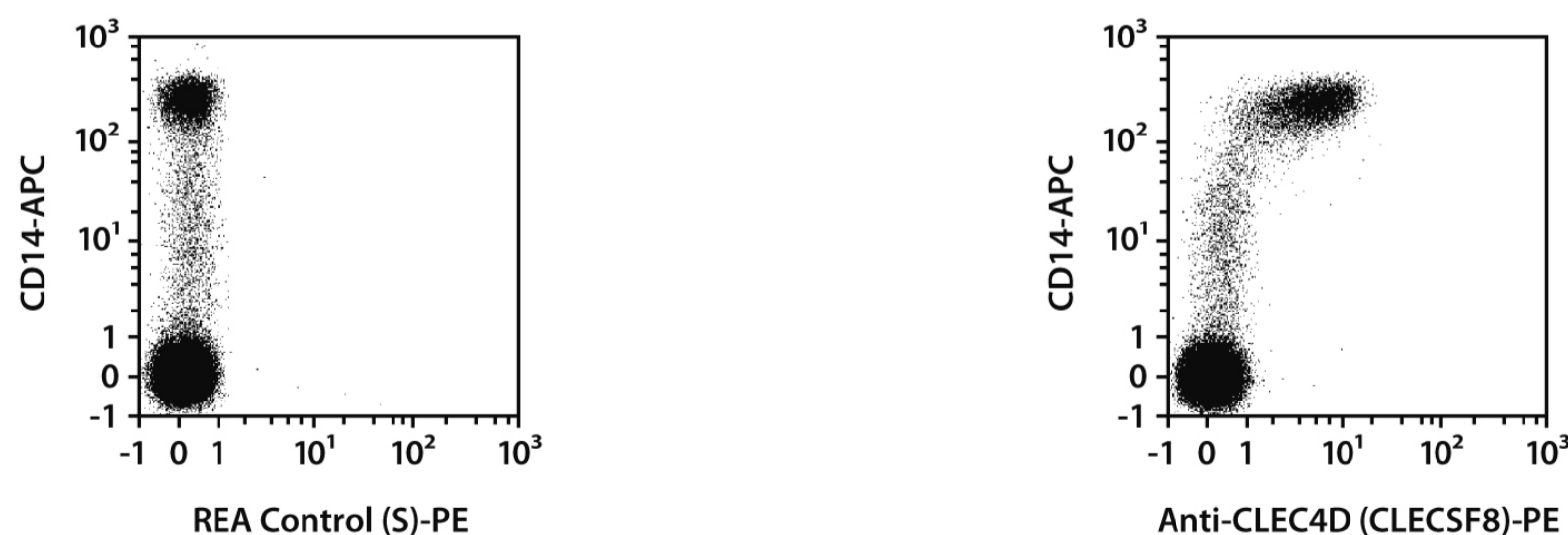
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS<sup>®</sup> BSA Stock Solution (# 130-091-376) 1:20 with autoMACS<sup>®</sup> Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).  
Note: EDTA can 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) 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.

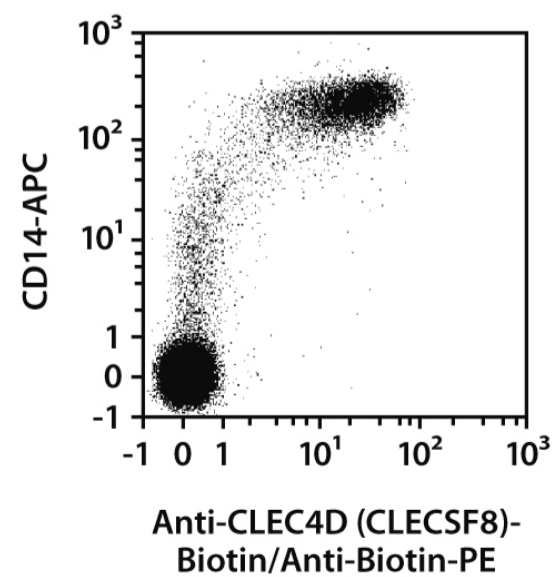
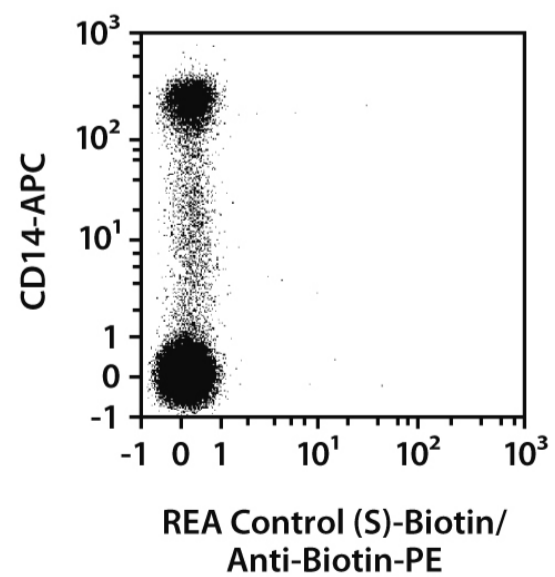
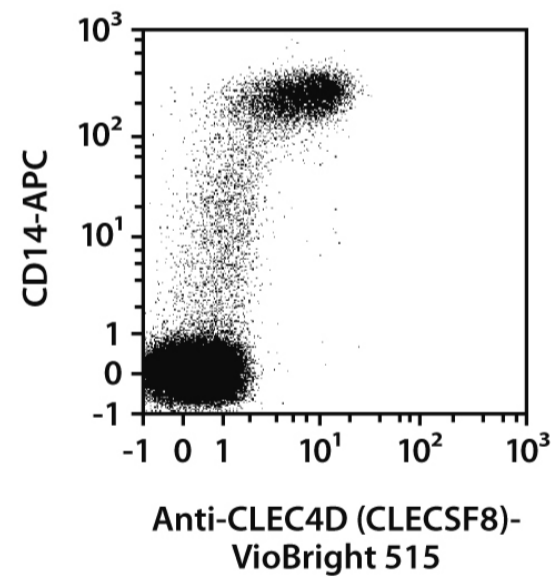
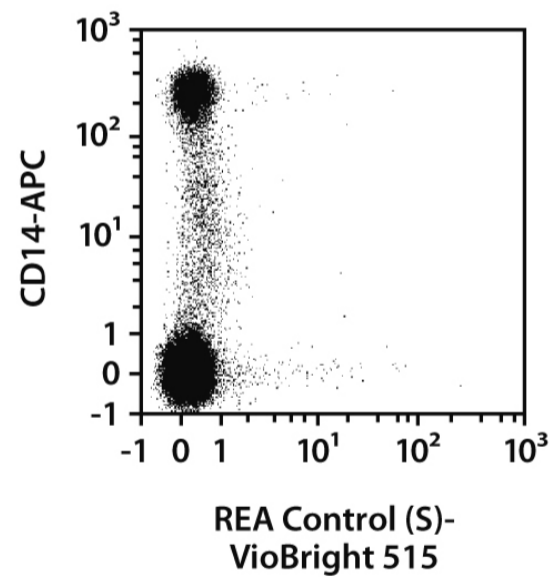
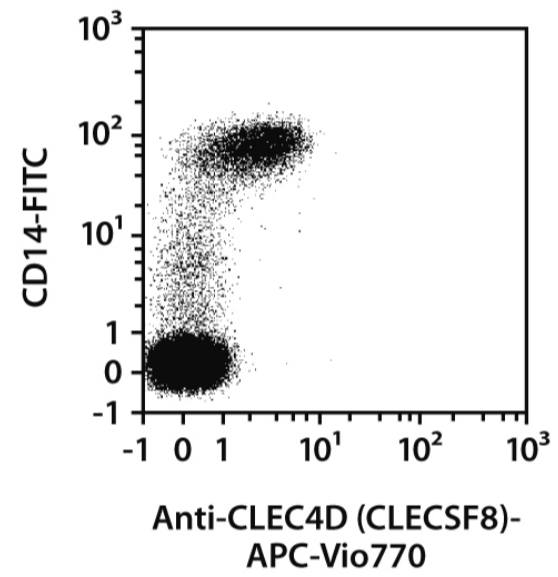
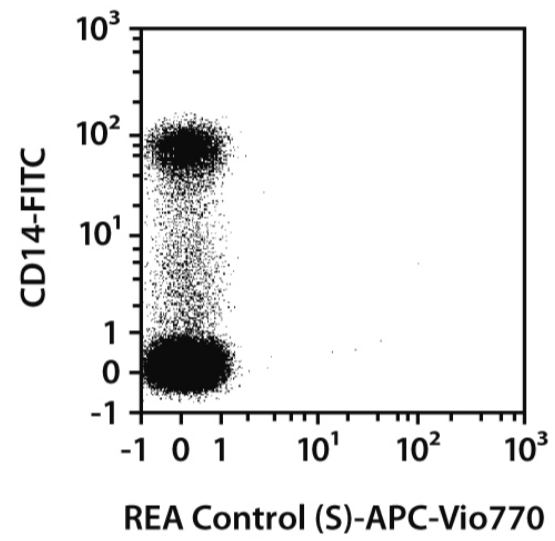
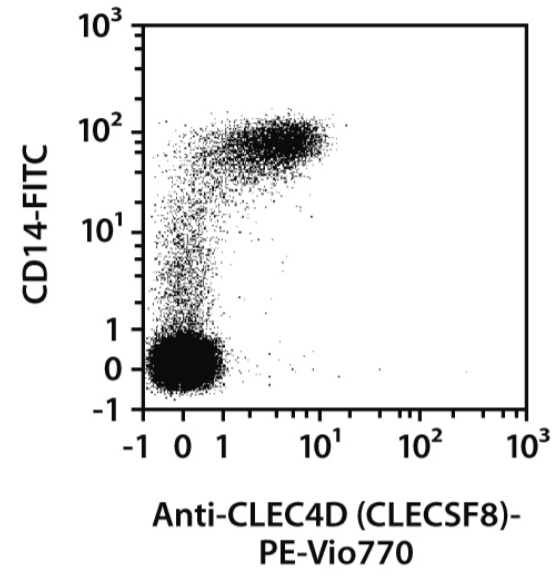
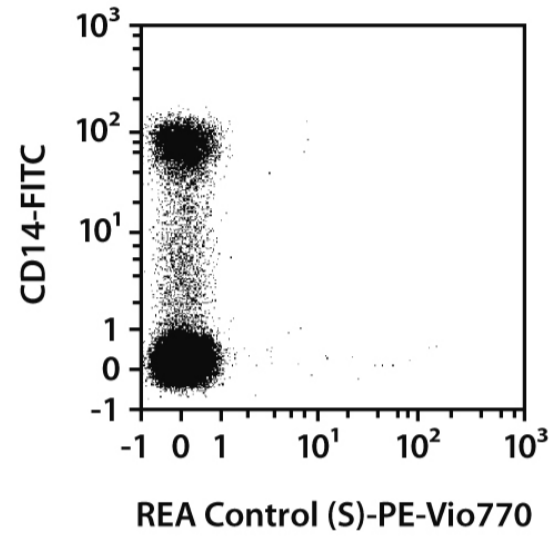
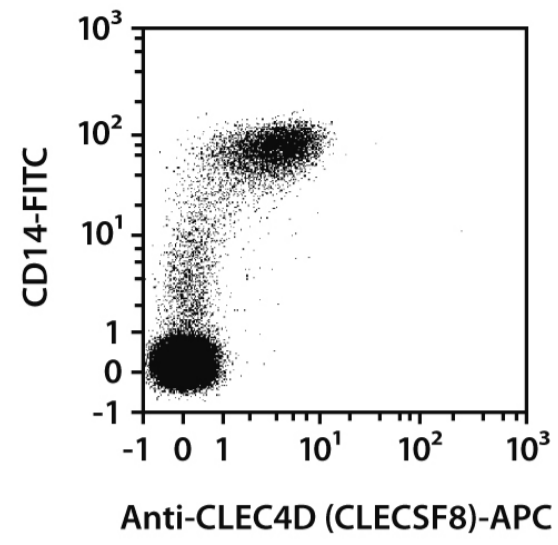
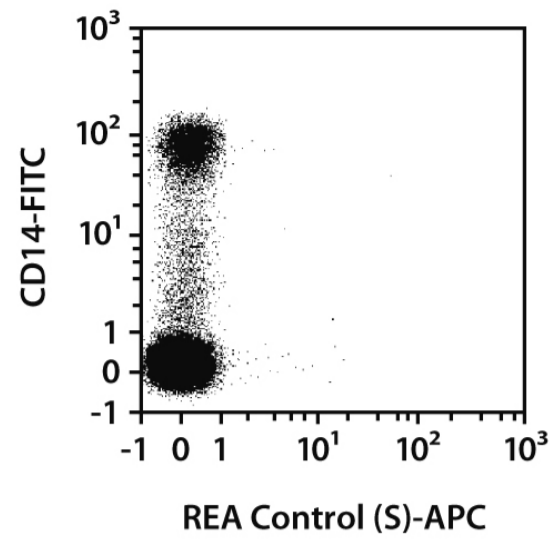
## Protocol for cell surface staining

- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:50 for up to 10<sup>6</sup> cells/100  $\mu$ L.
  - Volumes given below are for up to 10<sup>6</sup> nucleated cells. When working with fewer than 10<sup>6</sup> 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 $\times$ g for 10 minutes. Aspirate supernatant completely.
  3. Resuspend up to 10<sup>6</sup> nucleated cells per 98  $\mu$ L of buffer.
  4. Add 2  $\mu$ L 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 $\times$ 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 Anti-CLEC4D (CLECSF8) antibodies or with the corresponding REA Control (S) antibodies (left image) as well as with CD14 antibodies. Flow cytometry was performed using the MACSQuant<sup>®</sup> 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.





## Warranty

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