



Miltenyi Biotec

# Anti-PEAR1 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-PEAR1-PE	for 100 tests	130-111-611
Anti-PEAR1-PE	for 30 tests	130-111-689
Anti-PEAR1-APC	for 30 tests	130-111-690
Anti-PEAR1-APC	for 100 tests	130-111-612
Anti-PEAR1-VioBright 515	for 30 tests	130-111-691
Anti-PEAR1-VioBright 515	for 100 tests	130-111-613
Anti-PEAR1-Biotin	for 30 tests	130-111-688
Anti-PEAR1-Biotin	for 100 tests	130-111-610

## 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>	PEAR1
<b>Clone</b>	REA790
<b>Isotype</b>	recombinant human IgG1
<b>Isotype control</b>	REA Control (S) antibodies
<b>Alternative names of antigen</b>	MEGF12
<b>Distribution of antigen</b>	endothelial cells, platelets, heart, kidney, skeletal muscle, pancreas, ovary, breast, lung, brain, megakaryocytes, osteoblasts, leukocytes, macrophages
<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 REA790 recognizes the human platelet endothelial aggregation receptor 1 (PEAR1) antigen, a type I transmembrane protein also known as MEGF12. PEAR1, an EGF-containing receptor and member of the MEGF family, is found in various tissues. High levels of PEAR1 are found in platelets and endothelial cells, it is weakly expressed in peripheral blood leukocytes and macrophages. PEAR1 becomes activated by platelet contact and is involved in agonist-induced platelet aggregation. Additional information: Clone REA790 displays negligible binding to Fc receptors.

## Reagent requirements

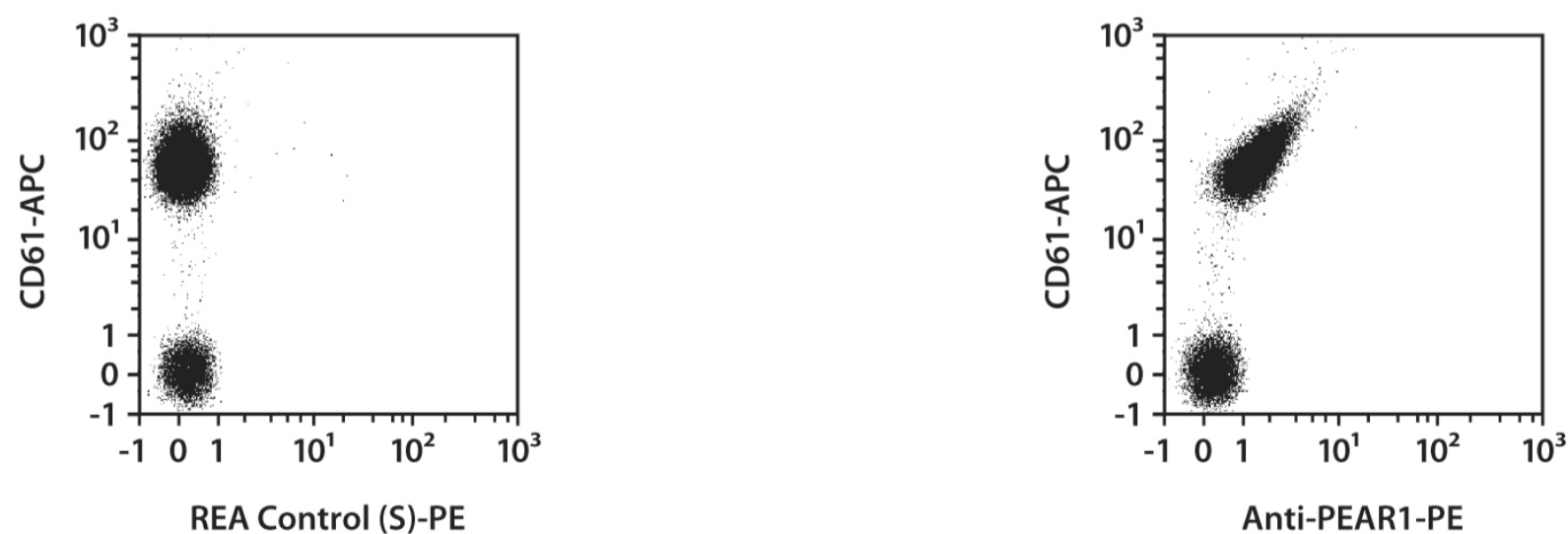
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS<sup>®</sup> BSA Stock Solution (# 130-091-376) 1:20 with autoMACS<sup>®</sup> Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).  
Note: EDTA can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

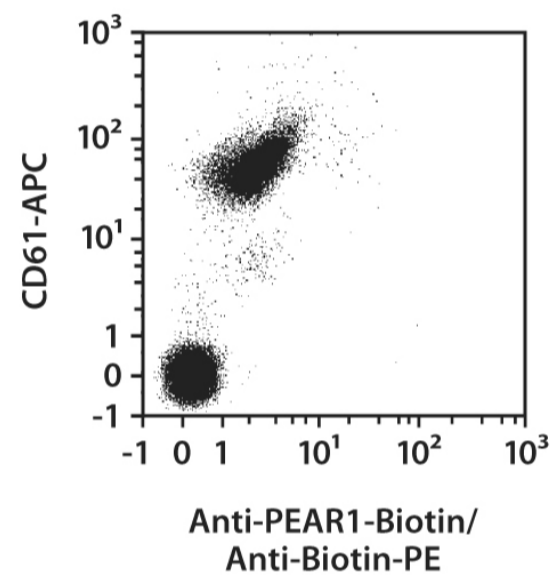
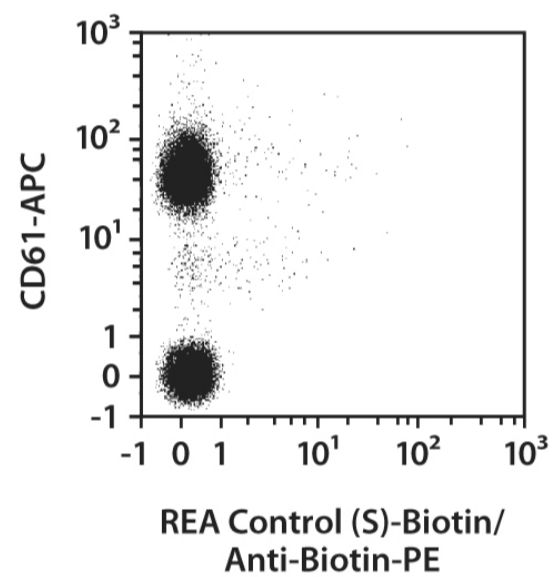
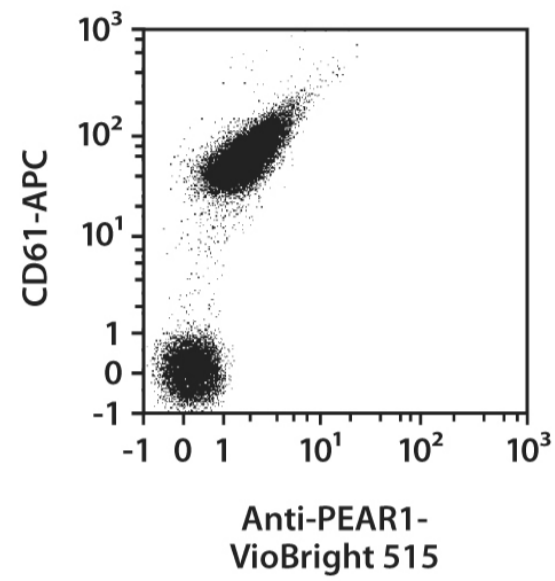
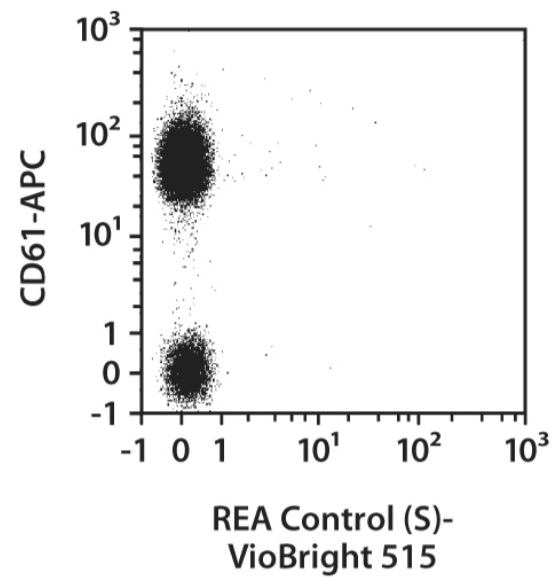
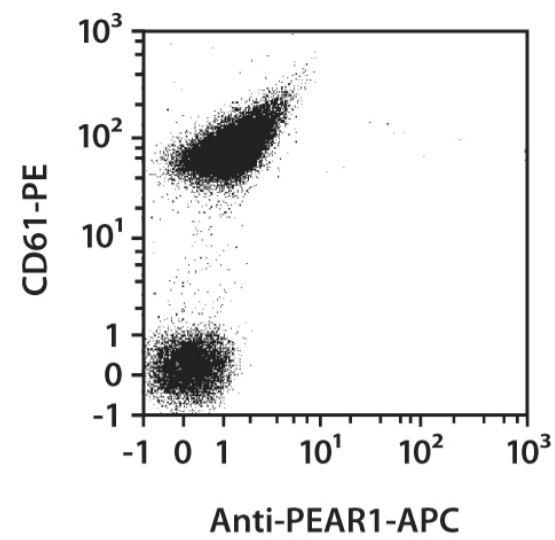
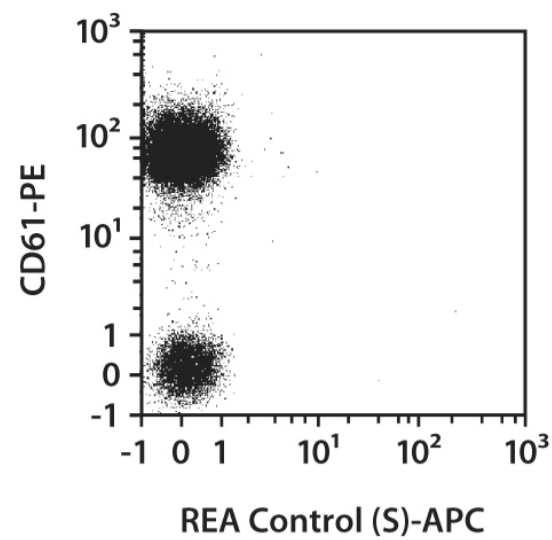
## Protocol for cell surface staining

- 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 µL.
  - 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×g for 10 minutes. Aspirate supernatant completely.
  3. Resuspend up to 10<sup>6</sup> 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

A mixture of human peripheral blood platelets and erythrocytes were stained with Anti-PEAR1 antibodies or with the corresponding REA Control (S) antibodies (left image) as well as with CD61 antibodies. Flow cytometry was performed using the MACSQuant<sup>®</sup> Analyzer. Cell debris and dead cells were excluded from the analysis based on scatter signals and propidium iodide.





## Warranty

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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](mailto:macs@miltenyibiotec.de) | [www.miltenyibiotec.com](http://www.miltenyibiotec.com) Miltenyi Biotec provides products and services worldwide. Visit [www.miltenyibiotec.com/local](http://www.miltenyibiotec.com/local) to find your nearest Miltenyi Biotec contact.

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