



Miltenyi Biotec

# CD45 antibodies, mouse

## For research use only

30 µg equal 100 tests, 150 µg equal 500 tests. One test corresponds to labeling of  $10^6$  cells.

Product	Content	Order no.
CD45-FITC	30 µg in 200 µL	130-116-500
CD45-FITC	150 µg in 1 mL	130-116-535
CD45-PE	30 µg in 200 µL	130-117-498
CD45-PE	150 µg in 1 mL	130-117-348
CD45-VioBlue	30 µg in 200 µL	130-119-130
CD45-VioBlue	150 µg in 1 mL	130-118-953
CD45-PE-Vio770	30 µg in 200 µL	130-117-529
CD45-APC-Vio770	30 µg in 200 µL	130-118-687
CD45-APC-Vio770	150 µg in 1 mL	130-118-559

## 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>	CD45
<b>Clone</b>	30F11
<b>Isotype</b>	rat IgG2bk
<b>Isotype control</b>	Rat IgG2b - isotype control antibodies
<b>Alternative names of antigen</b>	Ptprc, T200, Ly-5, LCA
<b>Entrez Gene ID</b>	<a href="#">19264</a>
<b>Molecular mass of antigen [kDa]</b>	142
<b>Distribution of antigen</b>	B cells, basophils, dendritic cells, granulocytes, hematopoietic stem cells, Langerhans cells, leukocytes, lymphocytes, macrophages, mast cells, monocytes, plasma cells, T cells, thymocytes
<b>Product format</b>	Reagents 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 30F11 recognizes the mouse CD45 antigen, also known as leukocyte common antigen (Ly-5), which is expressed at high levels on all cells of hematopoietic origin except for erythrocytes. Clone 30F11 reacts with all CD45 isoforms. CD45 can be used to discriminate leukocytes from non-hematopoietic cells.

## Reagent requirements

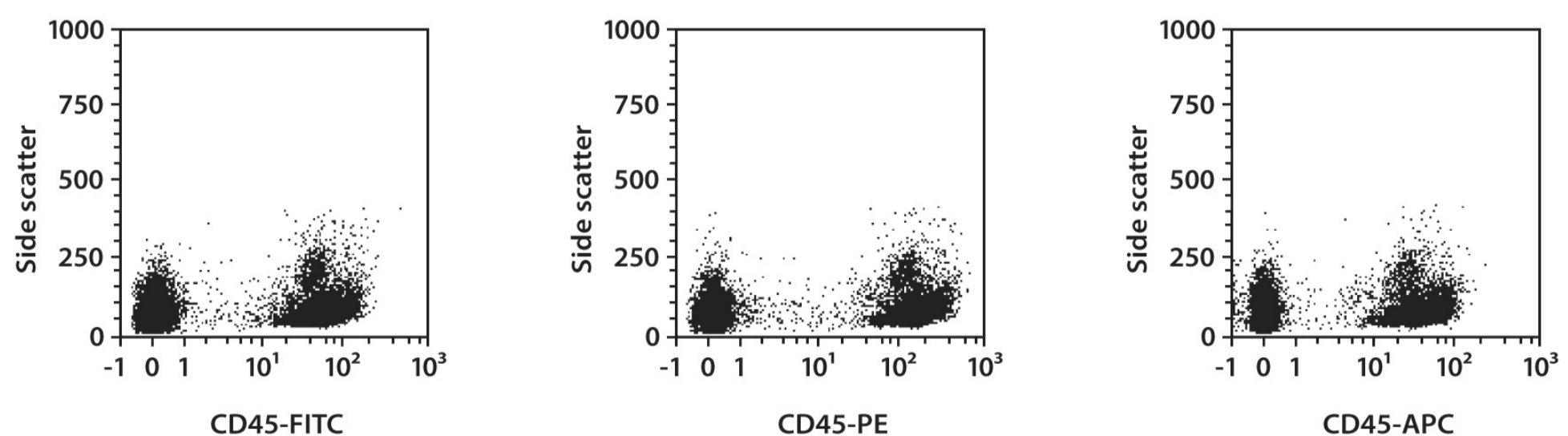
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS<sup>®</sup> BSA Stock Solution (# 130-091-376) 1:20 with autoMACS<sup>®</sup> Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).  
Note: EDTA can 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, mouse (# 130-092-575) 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.

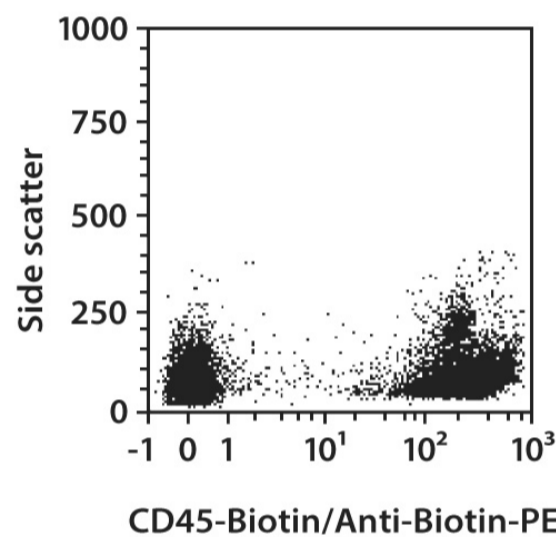
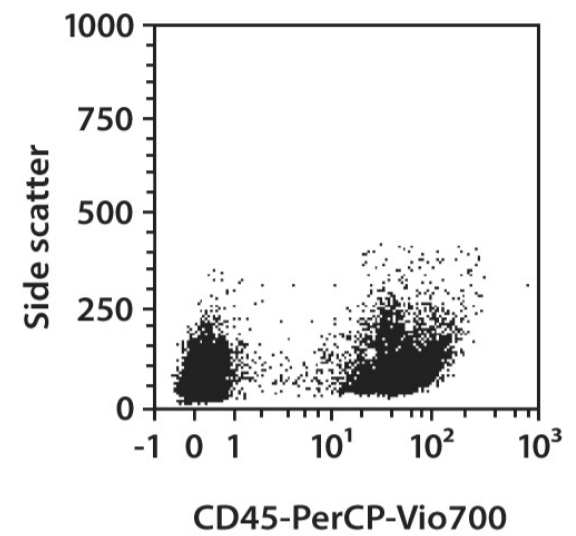
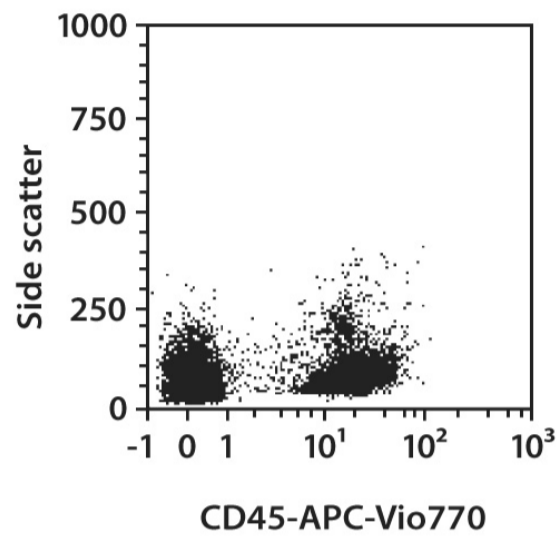
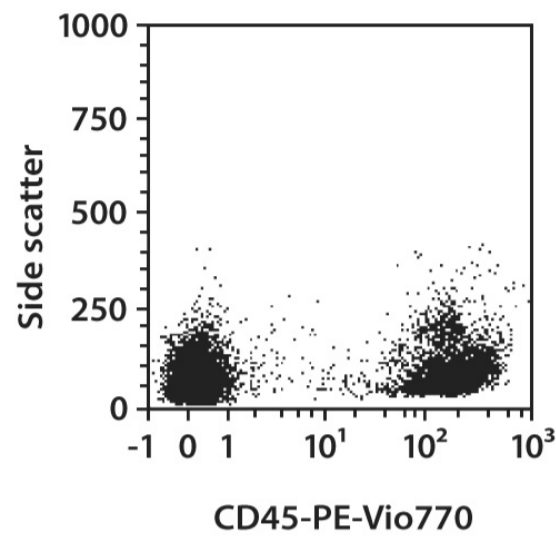
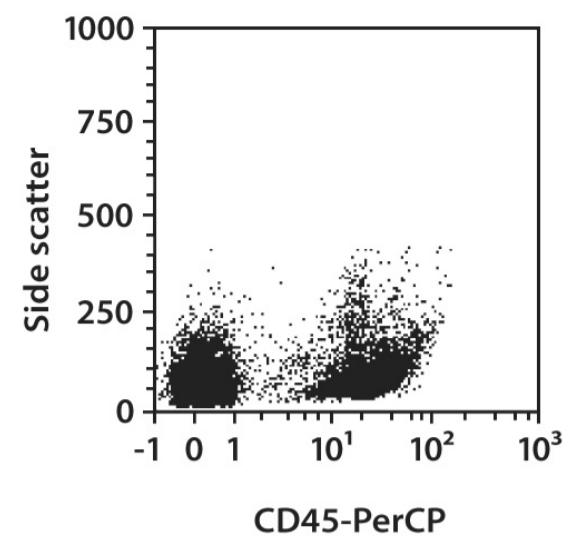
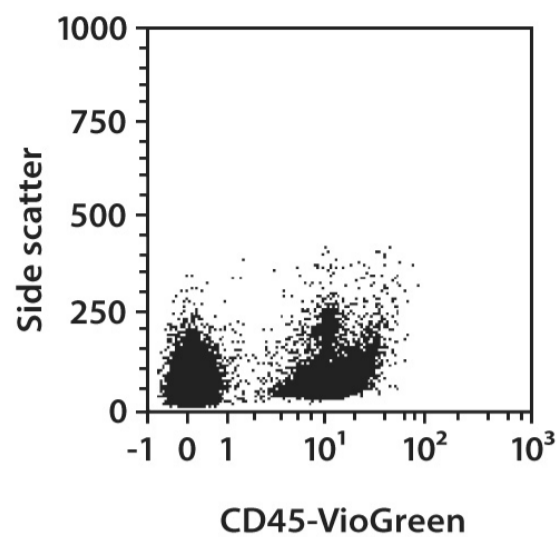
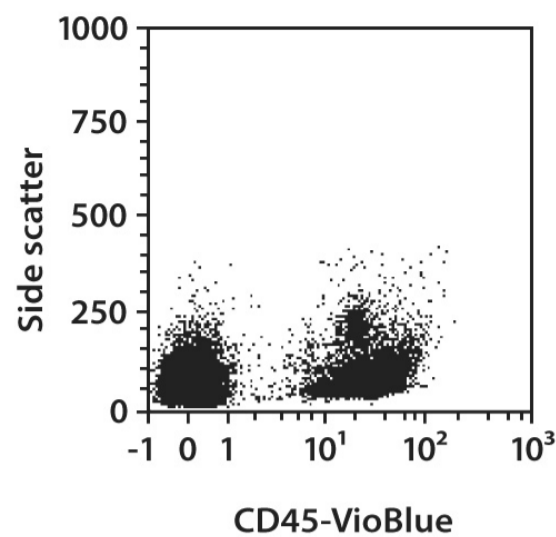
## Protocol for cell surface staining

- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:50 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

Mouse splenocytes were stained with CD45 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 fluorescence or 4',6-diamidino-2-phenylindole (DAPI) fluorescence, as in the case of tandem conjugates.





## Warranty

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