

CD117 antibodies, mouse

For research use only

30 μ g equal 100 tests, 150 μ g equal 500 tests. One test corresponds to labeling of 10[°] cells.

Product	Content	Order no.
CD117-Biotin	30 μg in 200 μL	130-111-692
CD117-PE	30 μg in 200 μL	130-111-693
CD117-PE	150 µg in 1 mL	130-111-615
CD117-APC	30 μg in 200 μL	130-111-694
CD117-PE-Vio770	30 μg in 200 μL	130-111-695
CD117-VioBright 515	30 μg in 200 μL	130-111-696
CD117-VioBright 515	150 µg in 1 mL	130-111-618
CD117-Biotin	150 µg in 1 mL	130-111-614

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	CD117
Clone	REA791
Isotype	recombinant human lgG1

Isotype control	REA Control antibodies
Alternative names of antigen	c-kit, SCO1, SCO5, SOW3, Ssm, Tr-kit, KIT, W, SCFR
Entrez Gene ID	<u>16590</u>
Molecular mass of antigen [kDa]	107
Distribution of antigen	B cells, bone marrow, brain, cancer stem cells, dendritic cells, endothelial cells, fibroblasts, hematopoietic stem cells, kidney, leukemia cells, liver, lung, mast cells, megakaryocytes, myeloid cells, NK cells, red blood 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 REA791 recognizes the mouse CD117 antigen, a 145 kDa cell surface glycoprotein belonging to the class III receptor tyrosine kinase family. CD117 is also known as c-kit or stem cell factor (SCF) receptor. It is expressed on the majority of hematopoietic progenitor cells including multipotent hematopoietic stem cells as well as some committed myeloid, erythroid and lymphoid precursor cells, and mature mast cells. CD117⁺ stem cells from murine bone marrow could also be differentiated into smooth muscle cells, myocytes, and endothelial cells*in vivo*.

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Additional information: Clone REA791 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 1:50 for up to 10° cells/100 µL.
- Volumes given below are for up to 10[°] nucleated cells. When working with fewer than 10[°] 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° nucleated cells per 98 µL of buffer.
- $4\cdot\,$ Add 2 μL of the antibody.
- 5. Mix well and incubate for 10 minutes in the dark in the refrigerator (2–8 $^{\circ}$ C).

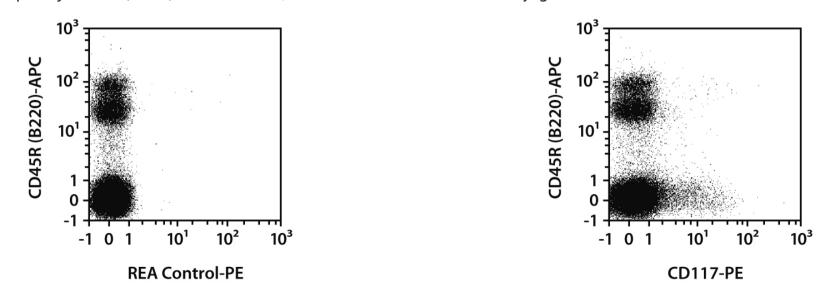
Note: Higher temperatures and/or longer incubation times may lead to non-specific cell labeling. Working on ice requires increased incubation times.

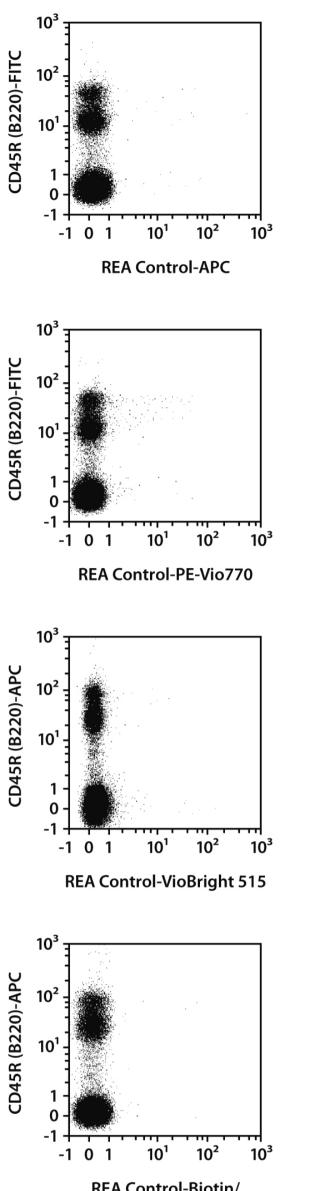
- 6. Wash cells by adding 1-2 mL of buffer and centrifuge at $300 \times g$ for 10 minutes. Aspirate supernatant completely.
- 7. (Optional) If biotinylated antibody was used, resuspend the cell pellet in buffer and stain with fluorochrome-conjugated antibiotin antibody according to the manufacturer's recommendations.
- 8. Resuspend cell pellet in a suitable amount of buffer for analysis by flow cytometry or fluorescence microscopy.

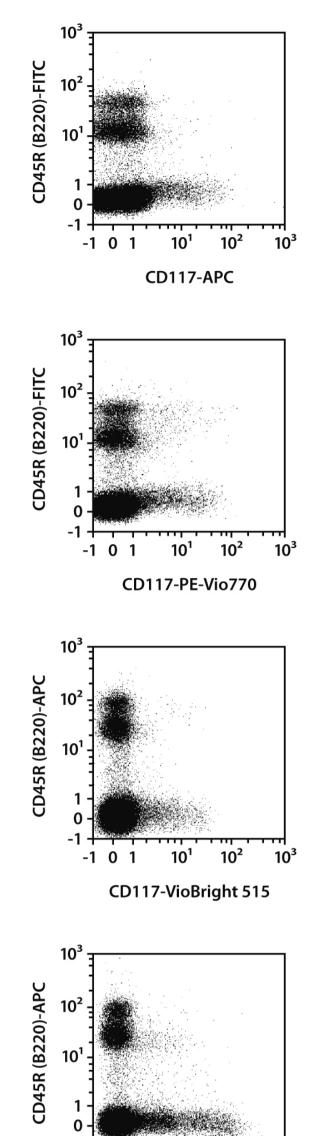
Examples of immunofluorescent staining

Bone marrow from C57BL/6 mice were stained with CD117 antibodies or with the corresponding REA Control antibodies (left image) as well as with CD45R (B220) antibodies. Flow cytometry was performed using the MACSQuant_®Analyzer. Cell debris

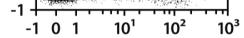
and dead cells were excluded from the analysis based on scatter signals and propidium iodide fluorescence or 4',6-diamidino-2-phenylindole (DAPI) fluorescence, as in the case of tandem conjugates.







REA Control-Biotin/ Anti-Biotin-PE



CD117-Biotin/Anti-Biotin-PE

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