CD45 antibodies, mouse

For research use only

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

Product	Content	Order no.
CD45-VioGreen	30 µg in 200 µL	130-110-803
CD45-FITC	30 µg in 200 µL	130-110-796
CD45-FITC	4x150 µg in 1 mL	130-110-658
CD45-PE	30 µg in 200 µL	130-110-797
CD45-PE	2x150 µg in 1 mL	130-110-659
CD45-APC	30 µg in 200 µL	130-110-798
CD45-APC	150 µg in 1 mL	130-110-660
CD45-VioBlue	30 µg in 200 µL	130-110-802
CD45-VioBlue	150 µg in 1 mL	130-110-664
CD45-VioGreen	150 µg in 1 mL	130-110-665
CD45-PE-Vio615	30 µg in 200 µL	130-110-804
CD45-PE-Vio615	150 µg in 1 mL	130-110-666
CD45-PE-Vio770	30 µg in 200 µL	130-110-799
CD45-PE-Vio770	150 µg in 1 mL	130-110-661
CD45-APC-Vio770	30 µg in 200 µL	130-110-800
CD45-APC-Vio770	150 µg in 1 mL	130-110-662
CD45-PerCP-Vio700	30 µg in 200 µL	130-110-801
CD45-PerCP-Vio700	150 µg in 1 mL	130-110-663
CD45-Biotin	30 µg in 200 µL	130-110-795
CD45-Biotin	4x150 µg in 1 mL	130-110-657

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	CD45
Clone	REA737
Isotype	recombinant human IgG1
Isotype control	REA Control antibodies
Alternative names of antigen	L-CA, LCA, Ly-5, T200

Entrez Gene ID	19264
Molecular mass of antigen [kDa]	142
Distribution of antigen	T cells, B 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 REA737 recognizes the mouse CD45 antigen, also known as Ly-5 or leukocyte common antigen (LCA), which belongs to the protein tyrosine phosphatase (PTP) family. CD45 is a glycoprotein which is involved in T cell receptor and B cell receptor signal transduction. It is expressed at high levels on all cells of hematopoietic origin except for erythrocytes. Clone REA737 reacts with all CD45 isoforms. CD45 can be used to discriminate leukocytes from non-hematopoietic cells. Additional information: Clone REA737 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.

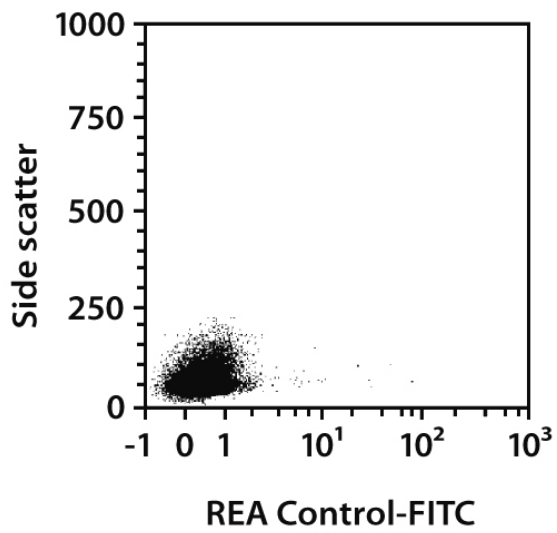
Protocol for cell surface staining

- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:50 for up to 10^6 cells/100 µL.
- Volumes given below are for up to 10^6 nucleated cells. When working with fewer than 10^6 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 10^6 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

Splenocytes from C57BL/6 mice were stained with CD45 antibodies or with the corresponding REA Control antibodies (left image). 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.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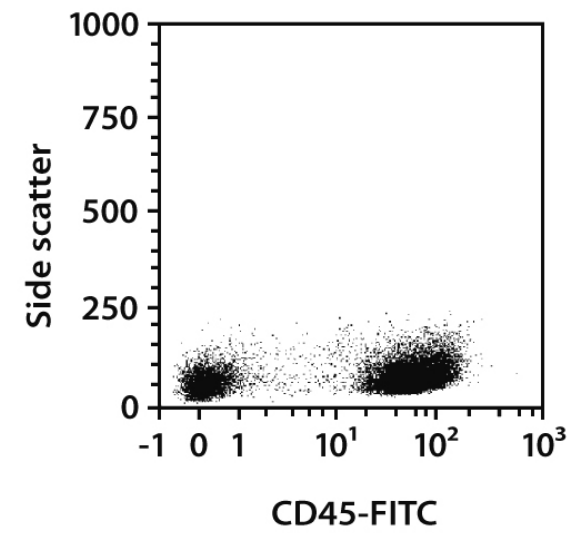
REA Control-FITC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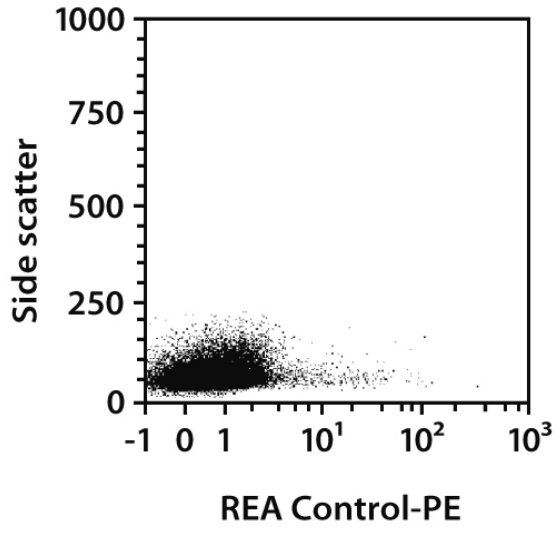
CD45-FITC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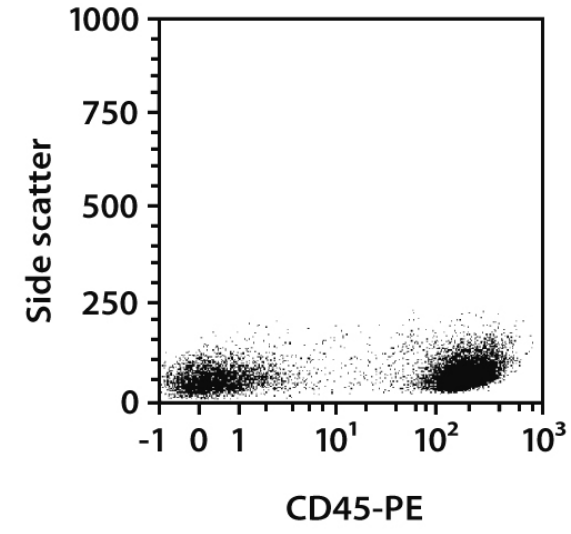
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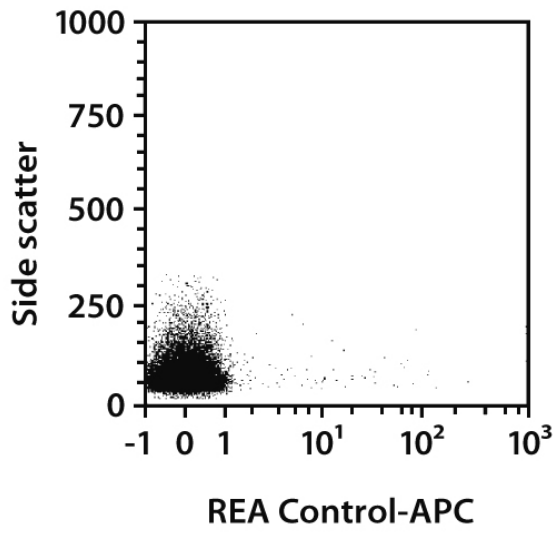
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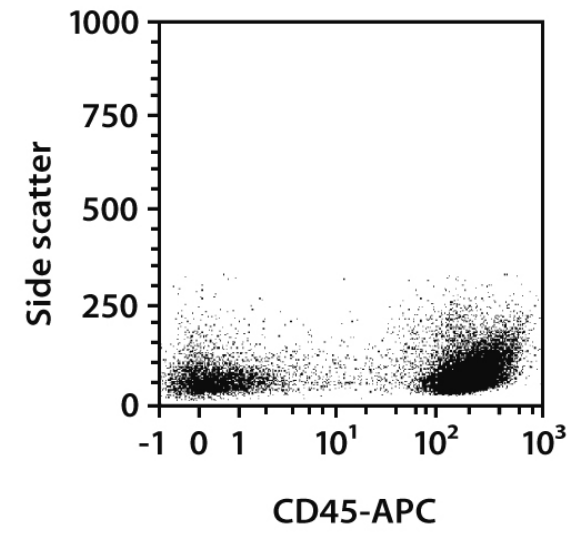
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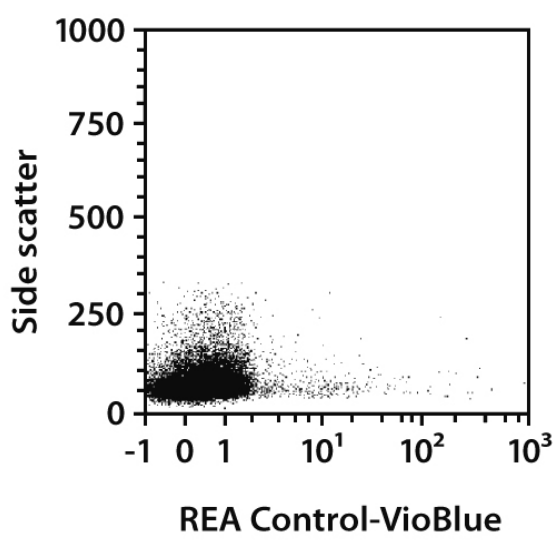
CD45-APC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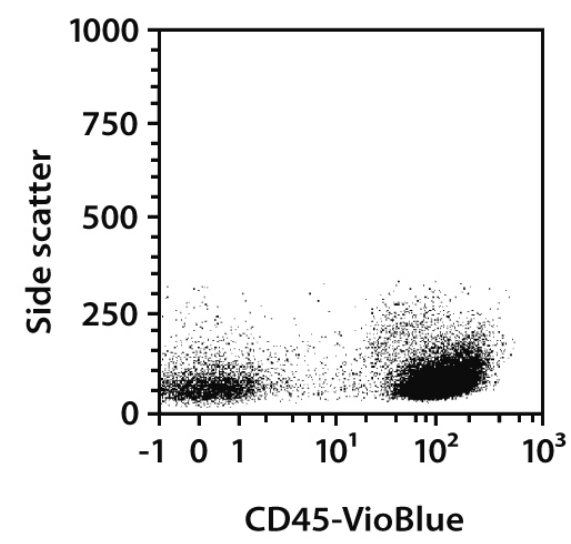
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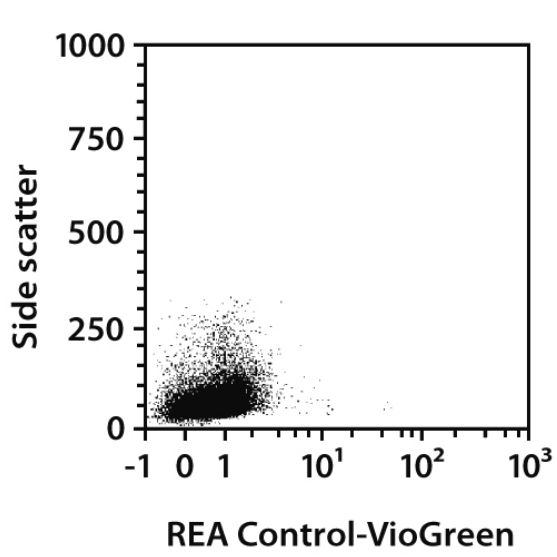
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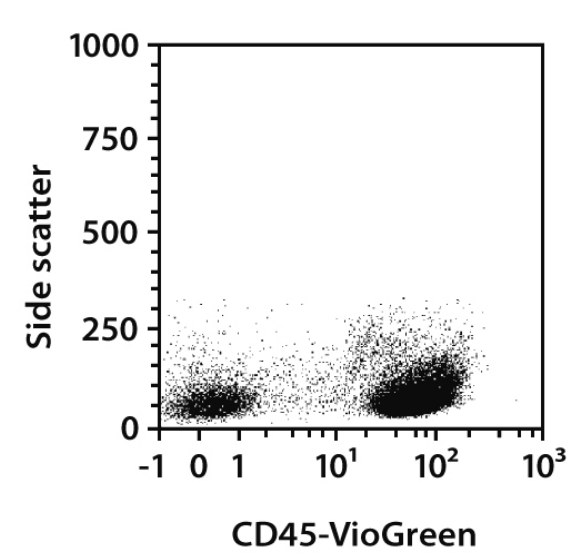
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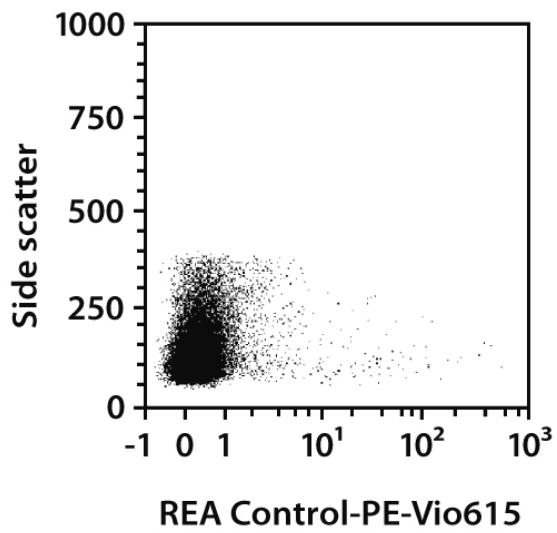
CD45-VioGreen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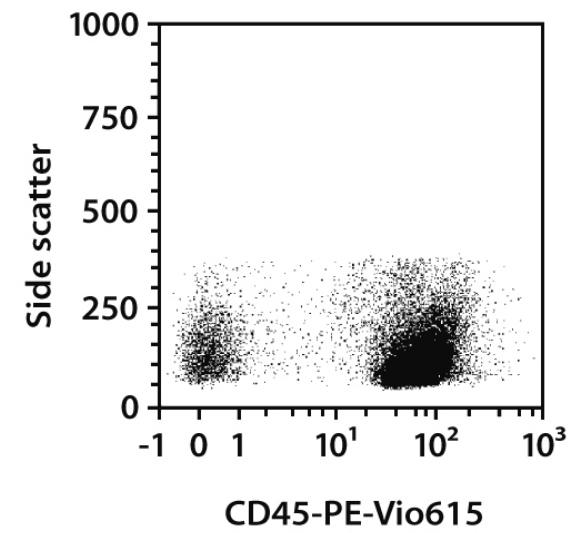
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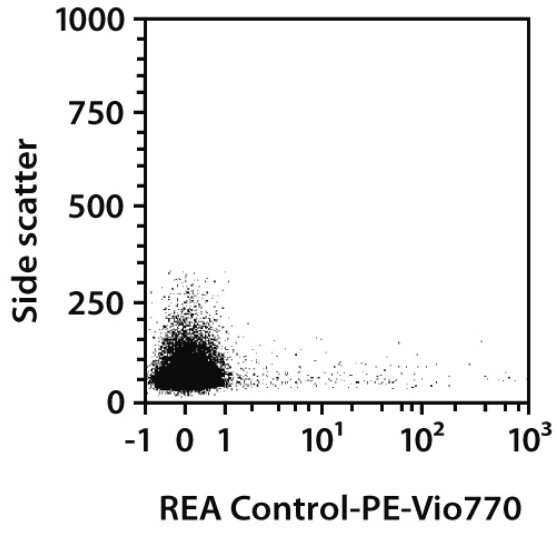
CD45-PE-Vio615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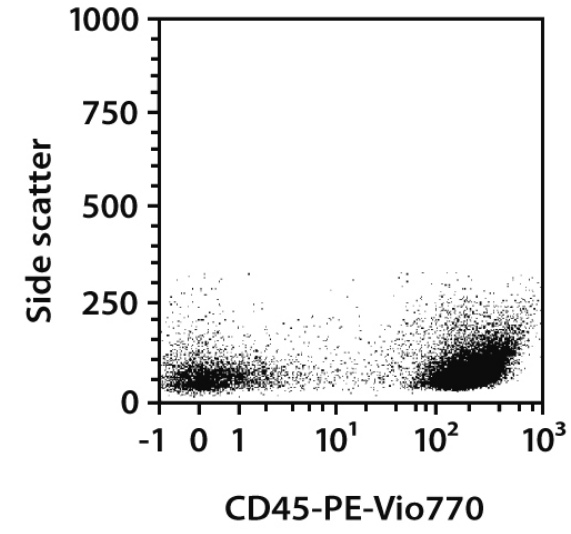
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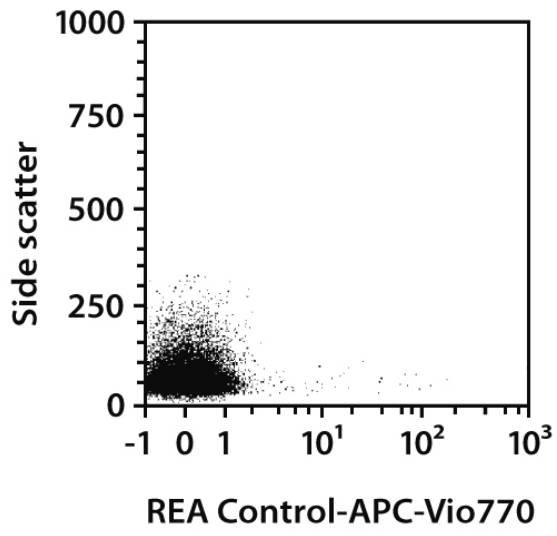
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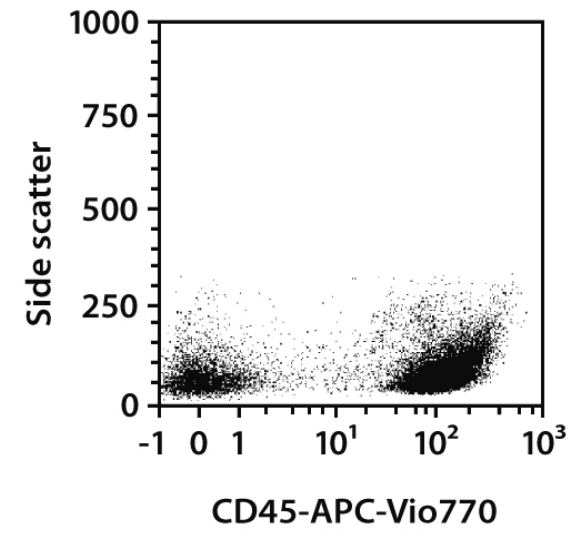
REA Control-APC-Vio770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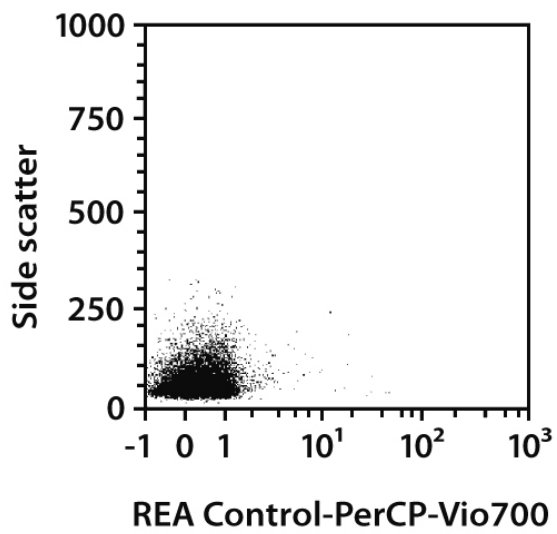
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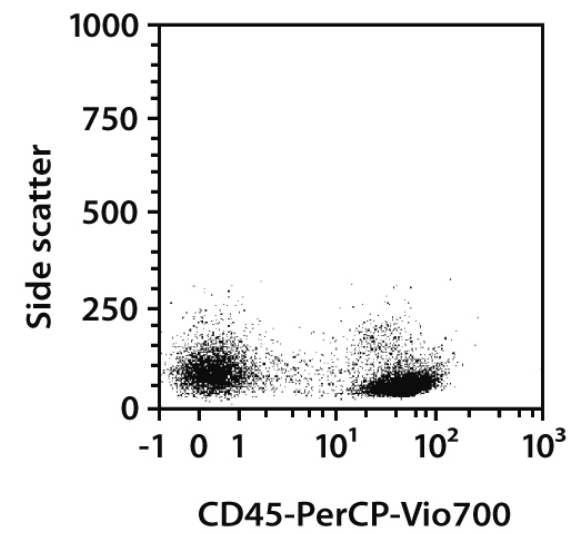
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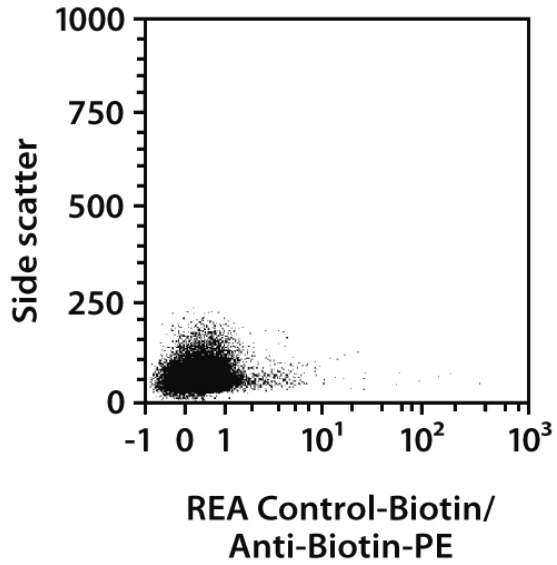
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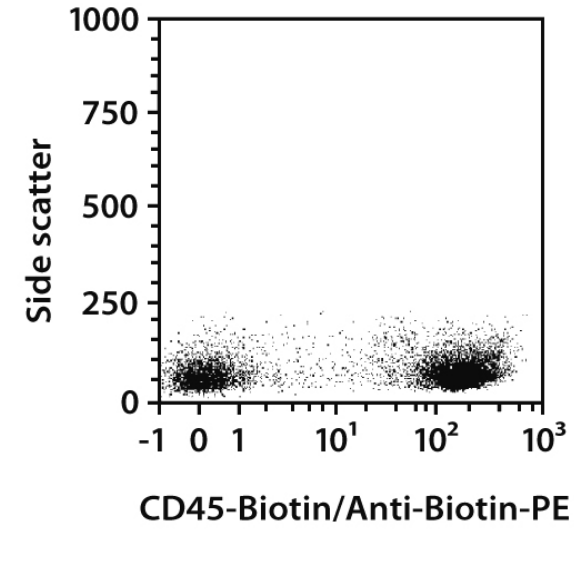
REA Control-Biotin/
Anti-Biotin-PE


Side scatter
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CD45-Biotin/Anti-Biotin-PE

Warranty

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