

CD4 antibodies, human

For research use only

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

Product	Content	Order no.
CD4-APC	for 30 tests	130-113-772
CD4-FITC	for 30 tests	130-113-775
CD4-FITC	for 100 tests	130-113-213
CD4-FITC	for 500 tests	130-114-580
CD4-VioBright FITC	for 30 tests	130-113-782
CD4-VioBright FITC	for 100 tests	130-113-220
CD4-PE	for 30 tests	130-113-776
CD4-PE	for 100 tests	130-113-214
CD4-APC	for 100 tests	130-113-210
CD4-VioBlue	for 30 tests	130-113-781
CD4-VioBlue	for 100 tests	130-113-219
CD4-VioGreen	for 30 tests	130-113-783
CD4-VioGreen	for 100 tests	130-113-221
CD4-PerCP	for 30 tests	130-113-779
CD4-PerCP	for 100 tests	130-113-217
CD4-PE-Vio615	for 30 tests	130-113-777
CD4-PE-Vio615	for 100 tests	130-113-215
CD4-PE-Vio770	for 30 tests	130-113-778
CD4-PE-Vio770	for 100 tests	130-113-216
CD4-APC-Vio770	for 30 tests	130-113-773
CD4-APC-Vio770	for 100 tests	130-113-211
CD4-PerCP-Vio700	for 30 tests	130-113-780
CD4-PerCP-Vio700	for 100 tests	130-113-218
CD4-Biotin	for 30 tests	130-113-774
CD4-Biotin	for 100 tests	130-113-212

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	CD4
Clone	VIT4
Isotype	mouse IgG2ak
Isotype control	Mouse IgG2a – isotype control antibodies
Alternative names of antigen	CD4mut, L3T4, Leu-3, T4
Entrez Gene ID	920
Molecular mass of antigen [kDa]	48
Distribution of antigen	dendritic cells, granulocytes, Langerhans cells, lymphocytes, macrophages, monocytes, neutrophils, T cells, T helper cells, thymocytes
Product format	Reagents are supplied in buffer containing stabilizer and 0.05% sodium azide.
Fixation	With the exception of the VioBlue conjugate the antibody is suited for staining of formaldehyde-fixed cell. Cells should be stained prior to fixation.
Storage	Store protected from light at 2–8 °C. Do not freeze.

Clone VIT4 recognizes the human CD4 antigen which is highly expressed on human T helper cells and thymocytes, and at lower levels on monocytes and dendritic cells. It is responsible for the recognition of the MHC class II antigen. VIT4 can be used for evaluating the transfection efficiency of cells transfected with pMACS 4.1 or pMACS 4-IRES when using the MACSelect™ – 4 Transfected Cell Selection Kit.

Reagent requirements

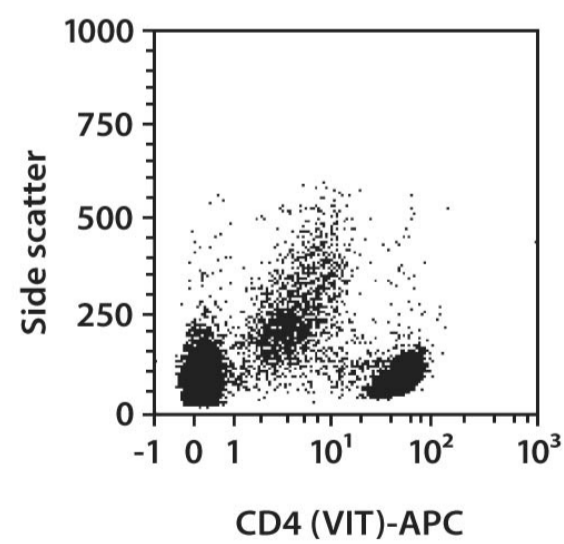
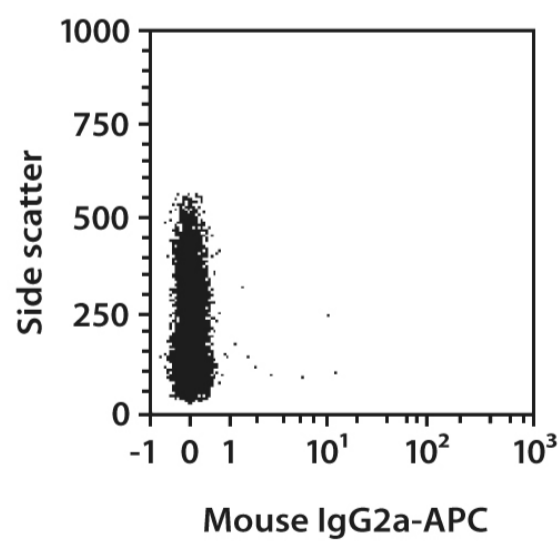
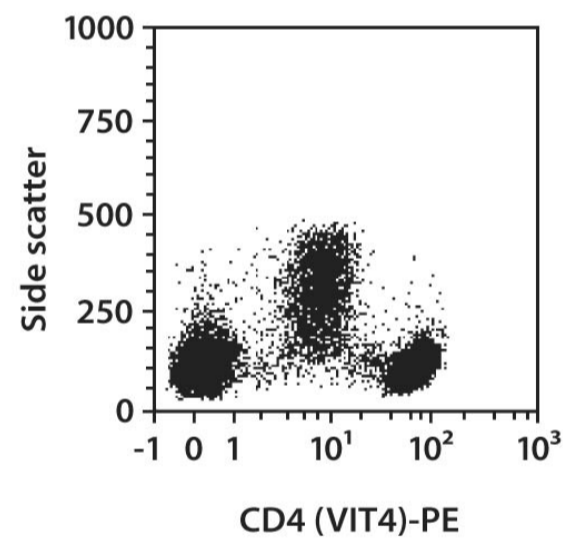
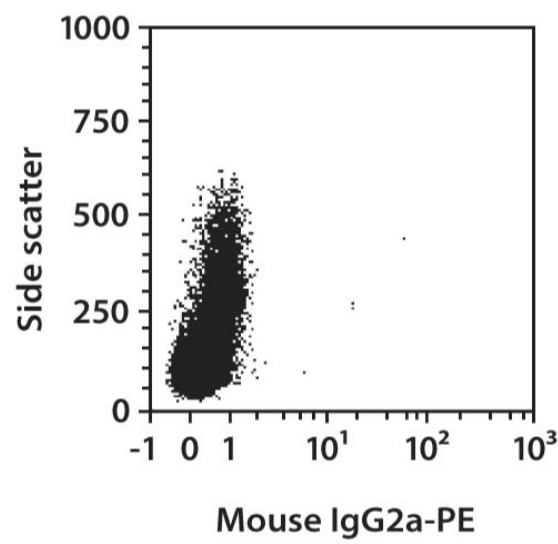
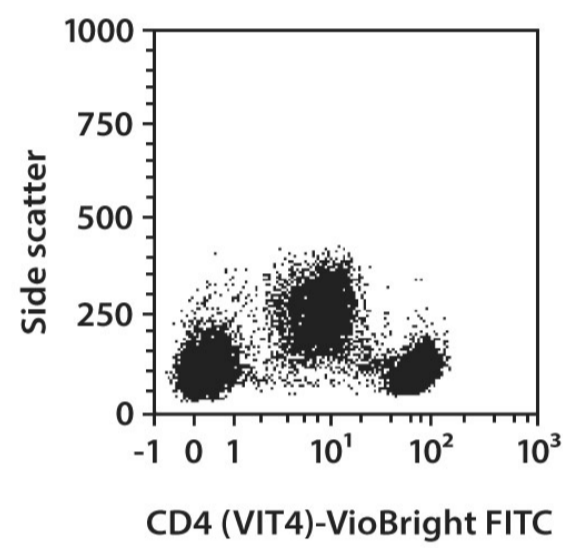
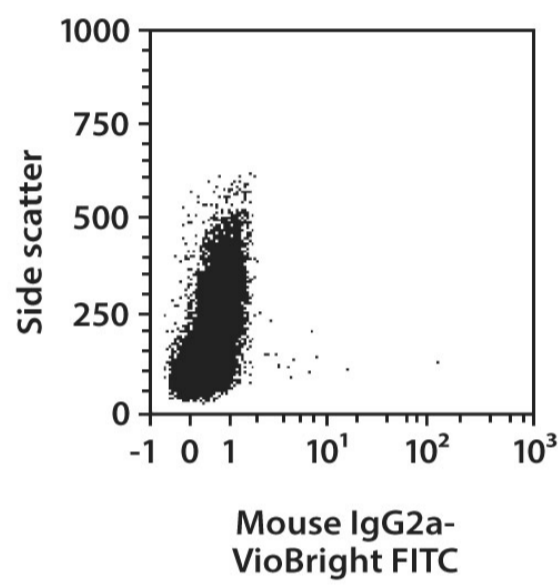
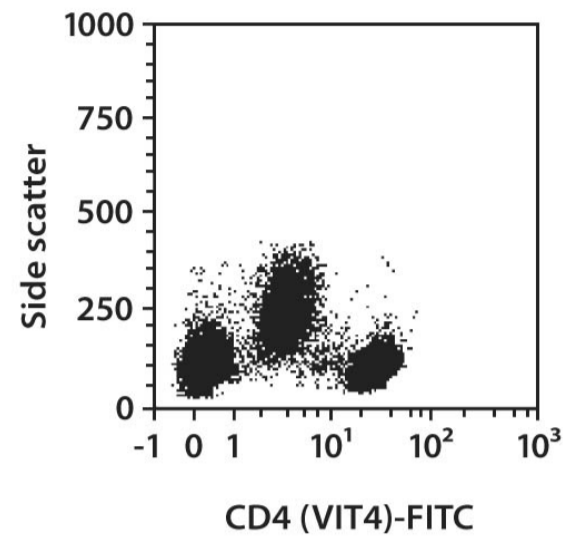
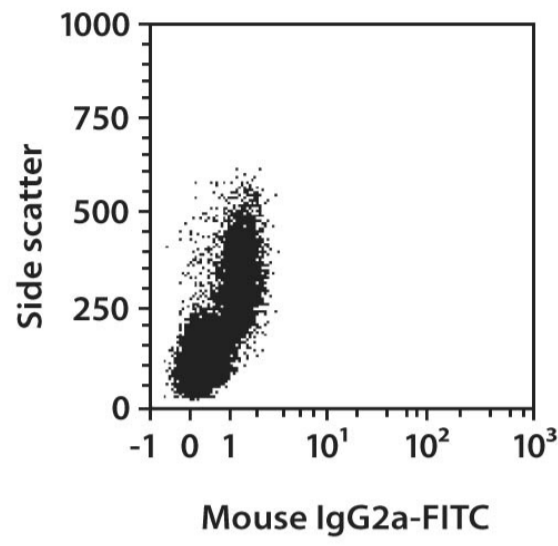
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS® BSA Stock Solution (# 130-091-376) 1:20 with autoMACS® Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).
Note: EDTA can be replaced by other supplements such as anticoagulant citrate dextrose formula-A (ACD-A) or citrate phosphate dextrose (CPD). Buffers or media containing Ca²⁺ or Mg²⁺ are not recommended for use.
- (Optional) FcR Blocking Reagent, human (# 130-059-901) 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.

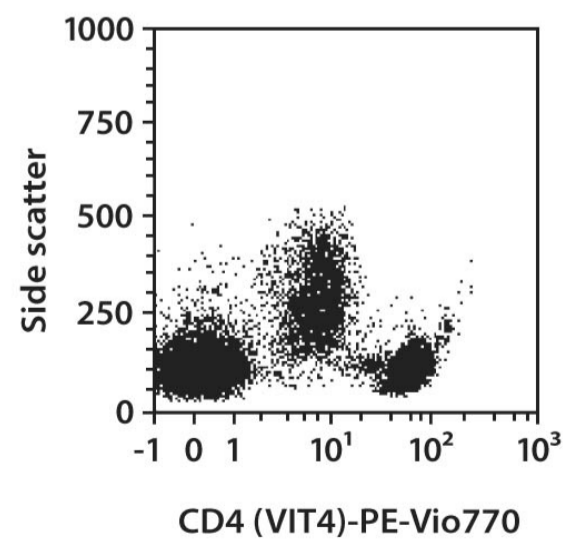
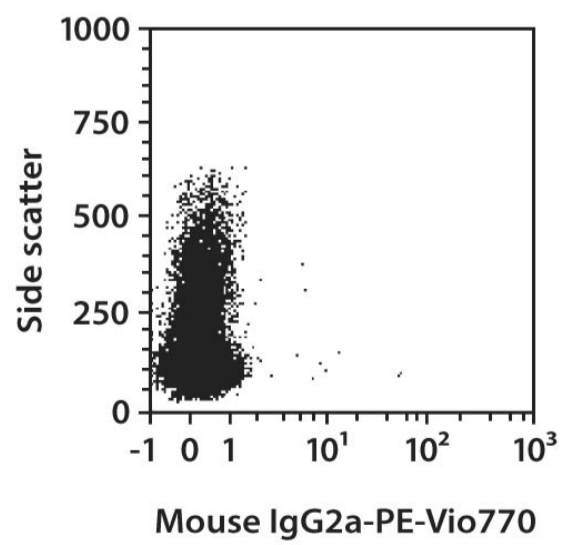
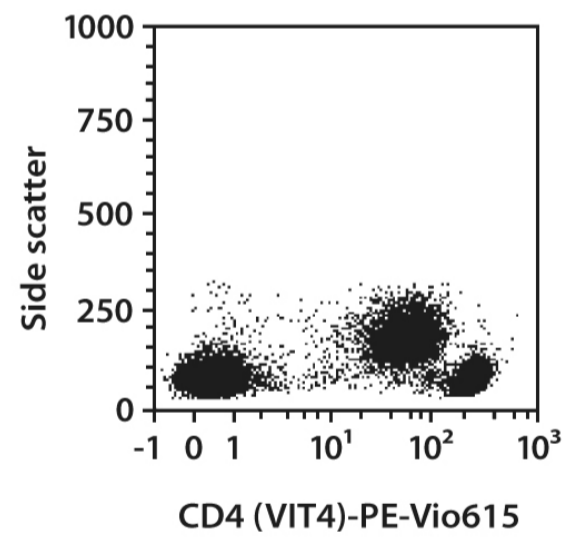
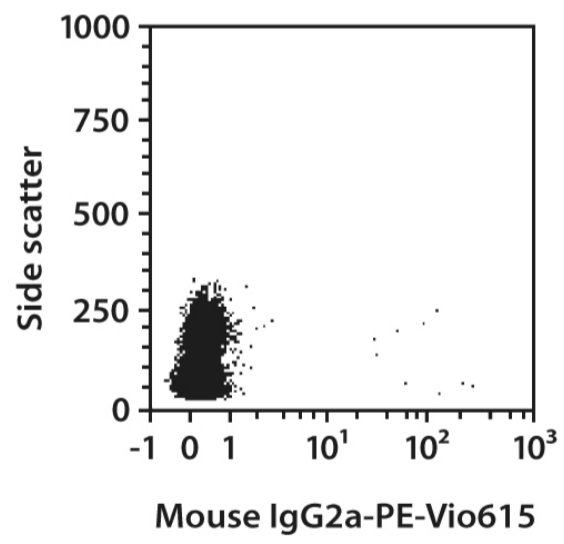
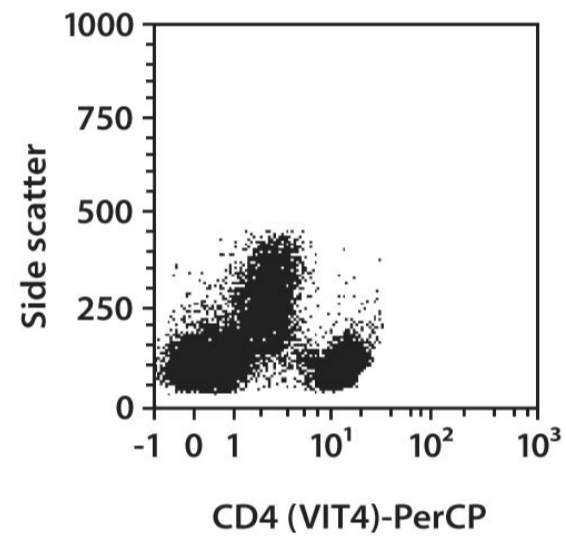
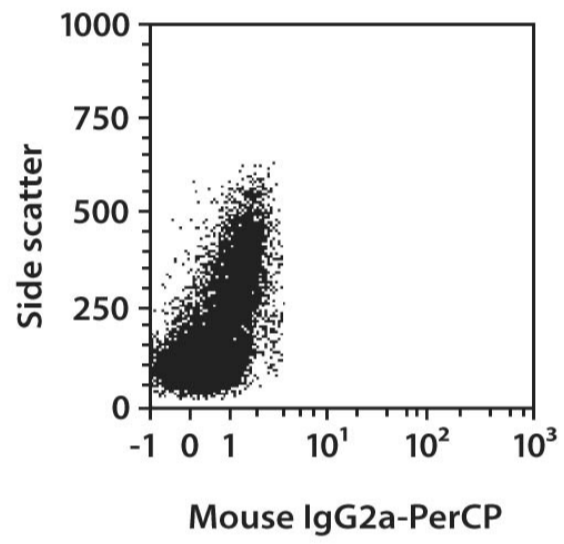
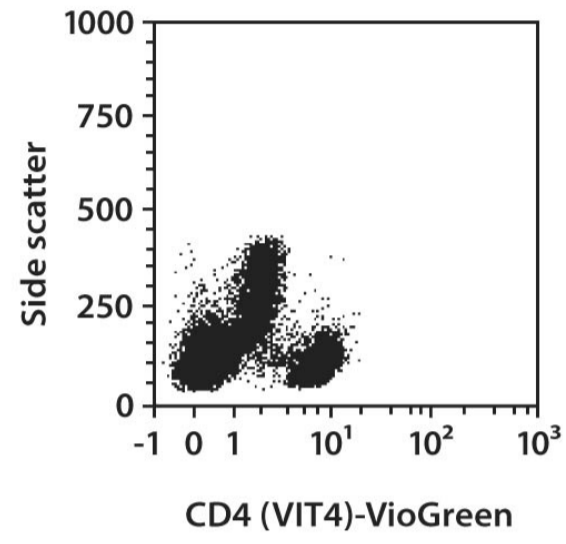
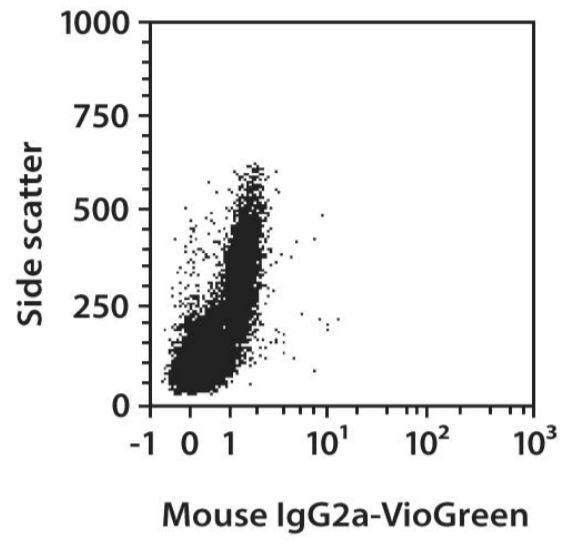
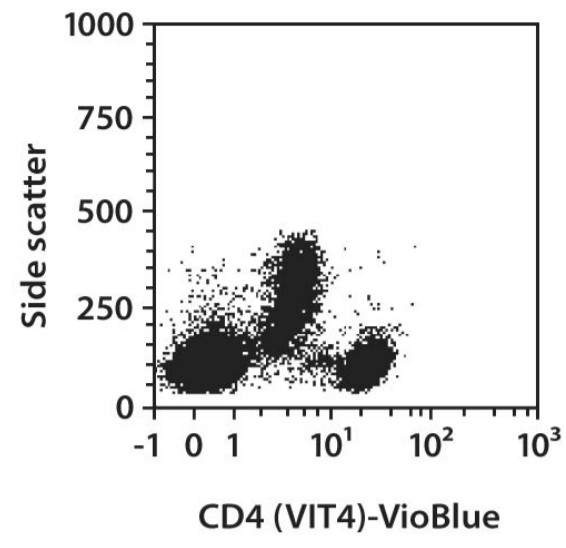
Protocol for cell surface staining

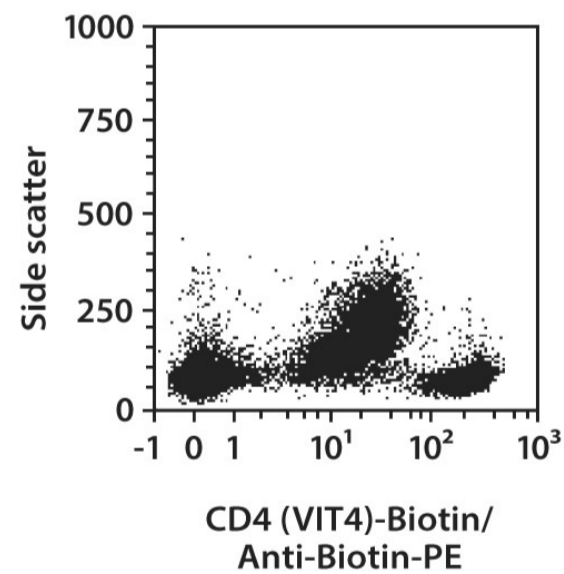
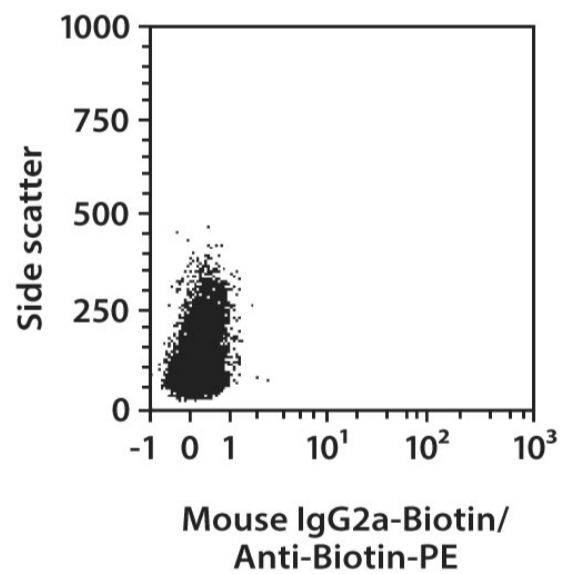
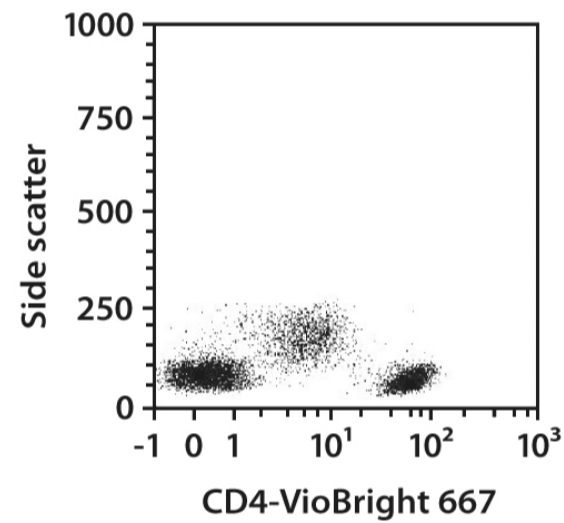
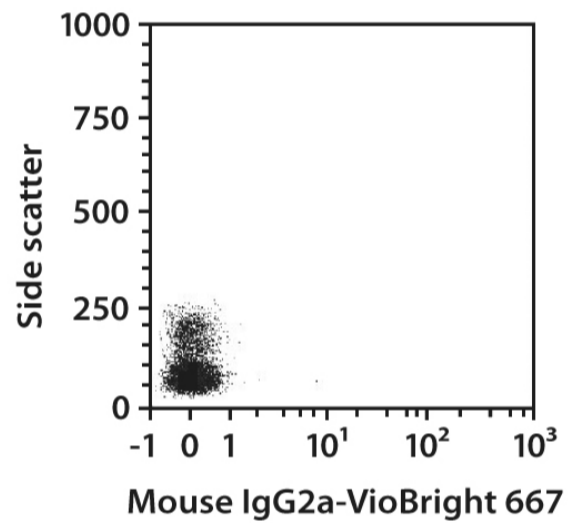
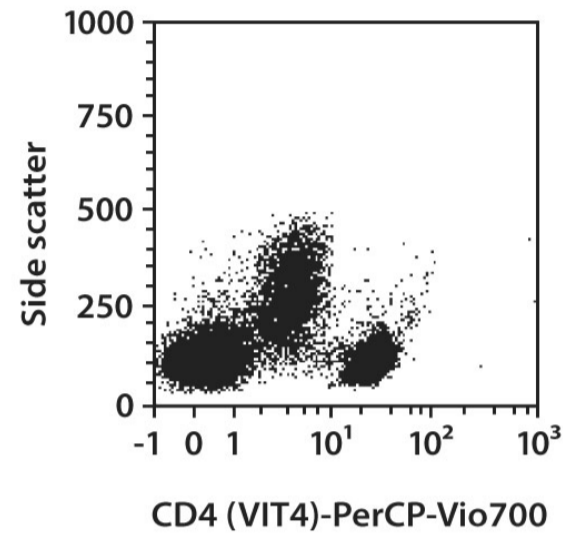
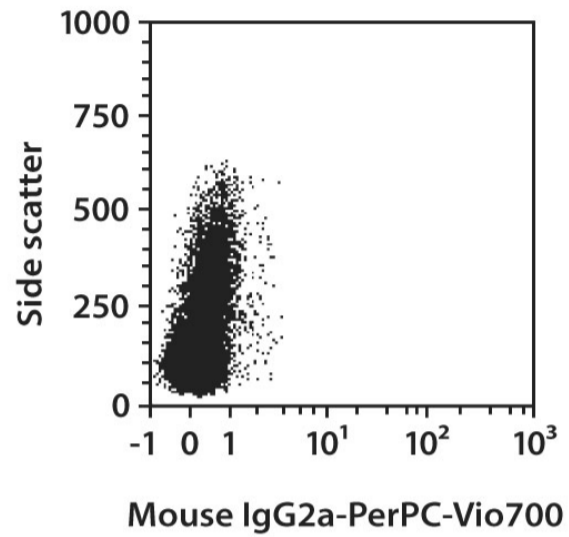
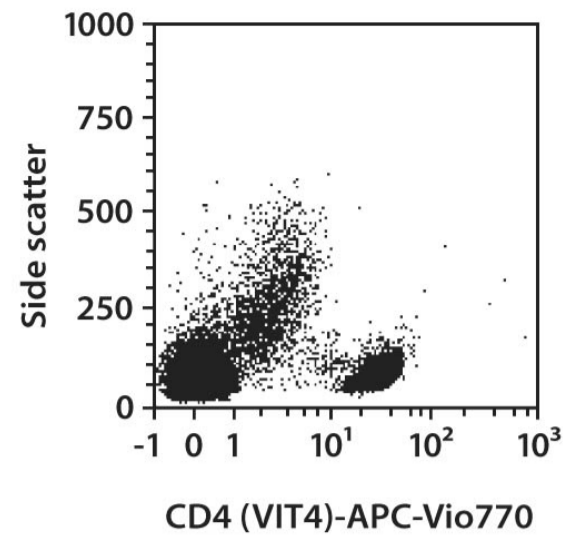
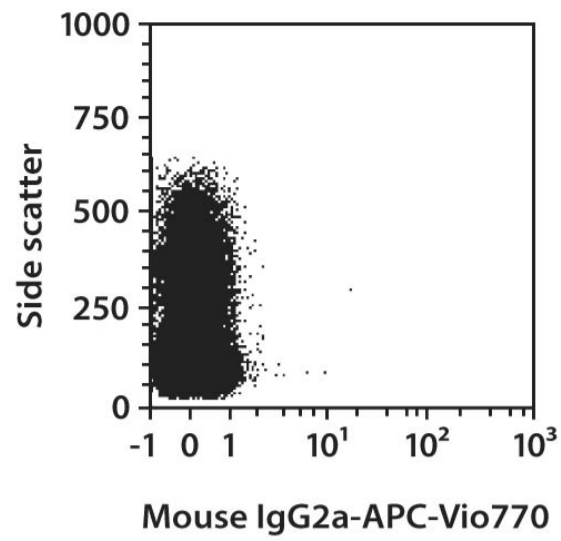
- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:50 for up to 10⁶ cells/100 µL.
 - Volumes given below are for up to 10⁶ nucleated cells. When working with fewer than 10⁶ cells, use the same volumes as indicated. When working with higher cell numbers, 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 10⁶ 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

Human peripheral blood mononuclear cells (PBMCs) were stained with CD4 antibodies or with the corresponding isotype control antibodies (left images) and analyzed by flow cytometry using the MACSQuant[®] 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

The products sold hereunder are warranted only to be free from defects in workmanship and material at the time of delivery to the customer. Miltenyi Biotec GmbH makes no warranty or representation, either expressed or implied, with respect to the fitness of a product for a particular purpose. There are no warranties, expressed or implied, which extend beyond the technical specifications of the products. Miltenyi Biotec GmbH's liability is limited to either replacement of the products or refund of the purchase price. Miltenyi Biotec GmbH is not liable for any property damage, personal injury or economic loss caused by the product.

macs@miltenyibiotec.de | www.miltenyibiotec.com Miltenyi Biotec provides products and services worldwide. Visit www.miltenyibiotec.com/local to find your nearest Miltenyi Biotec contact.

Unless otherwise specifically indicated, Miltenyi Biotec products and services are for research use only and not for therapeutic or diagnostic use. autoMACS, MACS, MACSQuant, Vio, VioBlue, VioBright, and VioGreen are either trademarks or registered trademarks of Miltenyi Biotec GmbH. Copyright © 2016 Miltenyi Biotec GmbH. All rights reserved.