



Miltenyi Biotec

CD14 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.
CD14-APC	for 30 tests	130-113-705
CD14-FITC	for 30 tests	130-113-708
CD14-FITC	for 100 tests	130-113-146
CD14-PE	for 30 tests	130-113-709
CD14-PE	for 100 tests	130-113-147
CD14-APC	for 100 tests	130-113-143
CD14-VioBlue	for 30 tests	130-113-714
CD14-VioBlue	for 100 tests	130-113-152
CD14-VioGreen	for 30 tests	130-113-715
CD14-VioGreen	for 100 tests	130-113-153
CD14-PerCP	for 30 tests	130-113-712
CD14-PerCP	for 100 tests	130-113-150
CD14-PE-Vio615	for 30 tests	130-113-710
CD14-PE-Vio615	for 100 tests	130-113-148
CD14-PE-Vio770	for 30 tests	130-113-711
CD14-PE-Vio770	for 100 tests	130-113-149
CD14-APC-Vio770	for 30 tests	130-113-706
CD14-APC-Vio770	for 100 tests	130-113-144
CD14-PerCP-Vio700	for 30 tests	130-113-713
CD14-PerCP-Vio700	for 100 tests	130-113-151
CD14-Biotin	for 30 tests	130-113-707
CD14-Biotin	for 100 tests	130-113-145

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	CD14
Clone	TÜK4
Isotype	mouse IgG2ak

Isotype control	Mouse IgG2a – isotype control antibodies
Alternative names of antigen	LPS R
Entrez Gene ID	929
Molecular mass of antigen [kDa]	37
Cross-reactivity	rhesus monkey (<i>Macaca mulatta</i>), cynomolgus monkey (<i>Macaca fascicularis</i>), pigtail monkey (<i>Macaca nemestrina</i>), cotton-top tamarin (<i>Saguinus oedipus</i>), pig, cow, sheep, goat, dog, mink, rabbit
Distribution of antigen	B cells, dendritic cells, epithelial cells, granulocytes, Langerhans cells, liver, macrophages, monocytes, osteoclasts, plasma 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 TÜK4 recognizes the human CD14 antigen and cross-reacts with non-human primate CD14. The CD14 antigen is a high affinity receptor for lipopolysaccharides (LPS) and LPS-binding protein (LBP)-complexes. It is part of the functional heteromeric LPS receptor complex comprised of CD14, TLR4, and MD-2. CD14 is strongly expressed on most human monocytes and macrophages in peripheral blood, other body fluids, and various tissues, such as lymph nodes and spleen. CD14 is expressed at high levels, also on a few CD1c (BDCA-1)⁺ CD2⁺ myeloid dendritic cells and at low levels on neutrophilic granulocytes. *Ex vivo* differentiation of monocytes to dendritic cells is associated with down-regulation of CD14 antigen expression.

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.

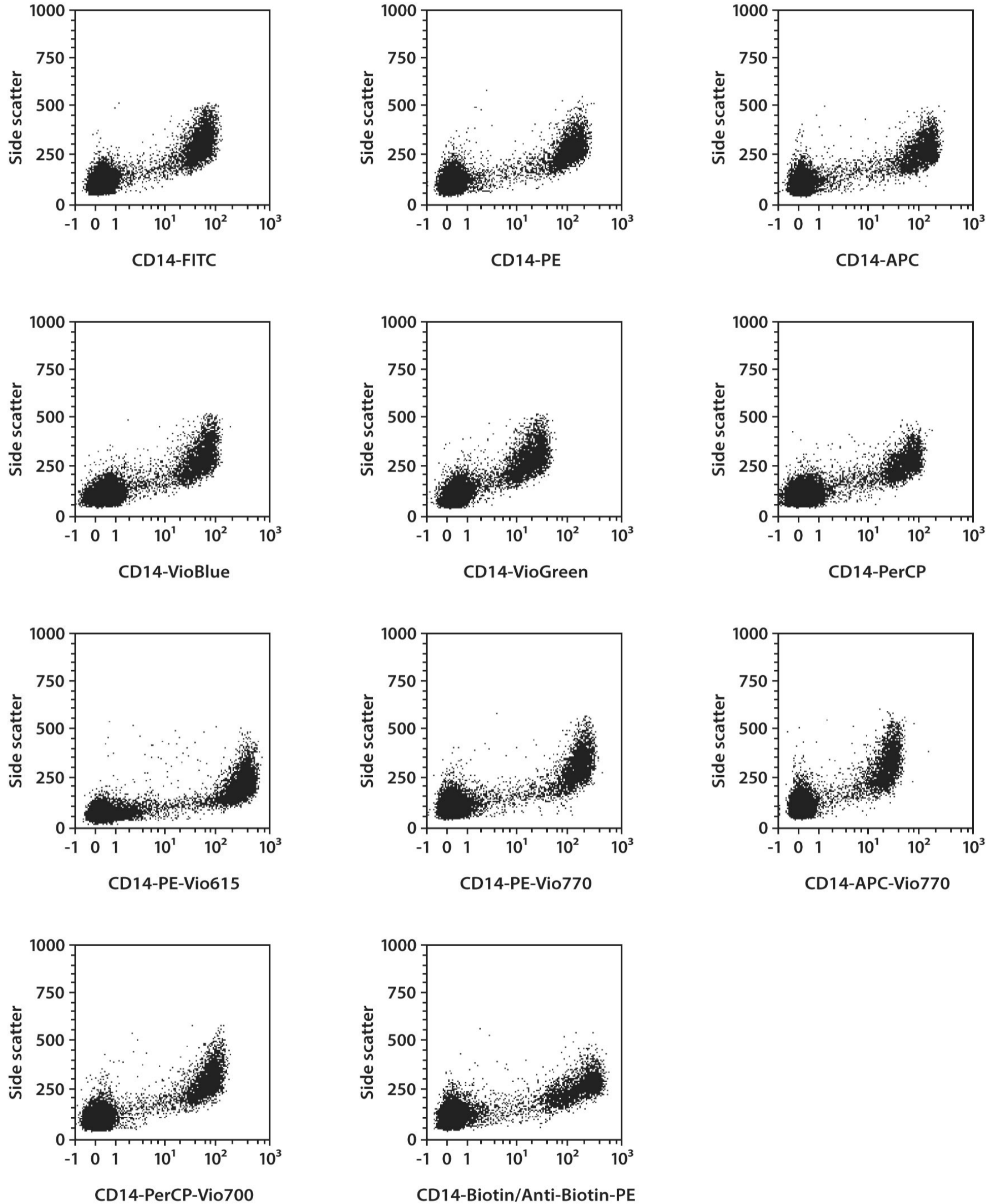
Protocol for cell surface staining

- 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 CD14 antibodies 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.

Miltenyi Biotec GmbH | Friedrich-Ebert-Straße 68 | 51429 Bergisch Gladbach | Germany | Phone +49 2204 8306-0 | Fax +49 2204 85197 | 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.