Anti-HLA-DR antibodies, human

For research use only

One test corresponds to labeling of up to 10^7 cells in a total volume of 100 µL.

Product	Content	Order no.
Anti-HLA-DR-FITC[1]	for 30 tests	130-098-176
Anti-HLA-DR-FITC[1]	for 100 tests	130-095-295
Anti-HLA-DR-PE	for 30 tests	130-098-177
Anti-HLA-DR-PE	for 100 tests	130-095-298
Anti-HLA-DR-APC	for 30 tests	130-098-178
Anti-HLA-DR-APC	for 100 tests	130-095-297
Anti-HLA-DR-VioBlue[1]	for 30 tests	130-098-175
Anti-HLA-DR-VioBlue[1]	for 100 tests	130-095-293
Anti-HLA-DR-PerCP[1]	for 30 tests	130-098-179
Anti-HLA-DR-PerCP[1]	for 100 tests	130-095-291
Anti-HLA-DR-PE-Vio770	for 30 tests	130-104-149
Anti-HLA-DR-PE-Vio770	for 100 tests	130-104-111
Anti-HLA-DR-APC-Vio770	for 30 tests	130-104-233
Anti-HLA-DR-APC-Vio770	for 100 tests	130-104-200
Anti-HLA-DR-PerCP-Vio700	for 30 tests	130-103-873
Anti-HLA-DR-PerCP-Vio700	for 100 tests	130-103-803
Anti-HLA-DR-Biotin[1]	for 30 tests	130-099-629
Anti-HLA-DR-Biotin[1]	for 100 tests	130-099-627

[1]Not recommended for cells that are labeled with MACS MicroBeads using the same antigen.

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	HLA-DR
Clone	AC122
Isotype	mouse IgG2aκ
Isotype control	Mouse IgG2a – isotype control antibodies
Alternative names of antigen	HLA-DRA, HLA-DRB1, HLA-DRB3, HLA-DRB4, HLA-DRB5
Molecular mass of antigen [kDa]	27-33
Cross-reactivity	rhesus monkey (*Macaca mulatta*), cynomolgus monkey (*Macaca fascicularis*), african green monkey (*Chlorocebus aethiops*), common marmoset (*Callithrix jacchus*), guinea pig

Product format	Antibodies are supplied in buffer containing stabilizer and 0.05% sodium azide.
Fixation	The antibody is suited for staining of formaldehyde-fixed cells.
Storage	Store protected from light at 2–8 °C. Do not freeze.

Anti-HLA-DR antibodies react with the human major histocompatibility (MHC) class II antigen HLA-DR. HLA-DR is constitutively expressed on professional antigen-presenting cells like dendritic cells, B cells, and monocytes/macrophages. Its expression is further up-regulated upon activation. On T cells, NK cells, hematopoietic precursor cells, and some epithelial cells the expression of HLA-DR is induced by cell activation.

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) 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

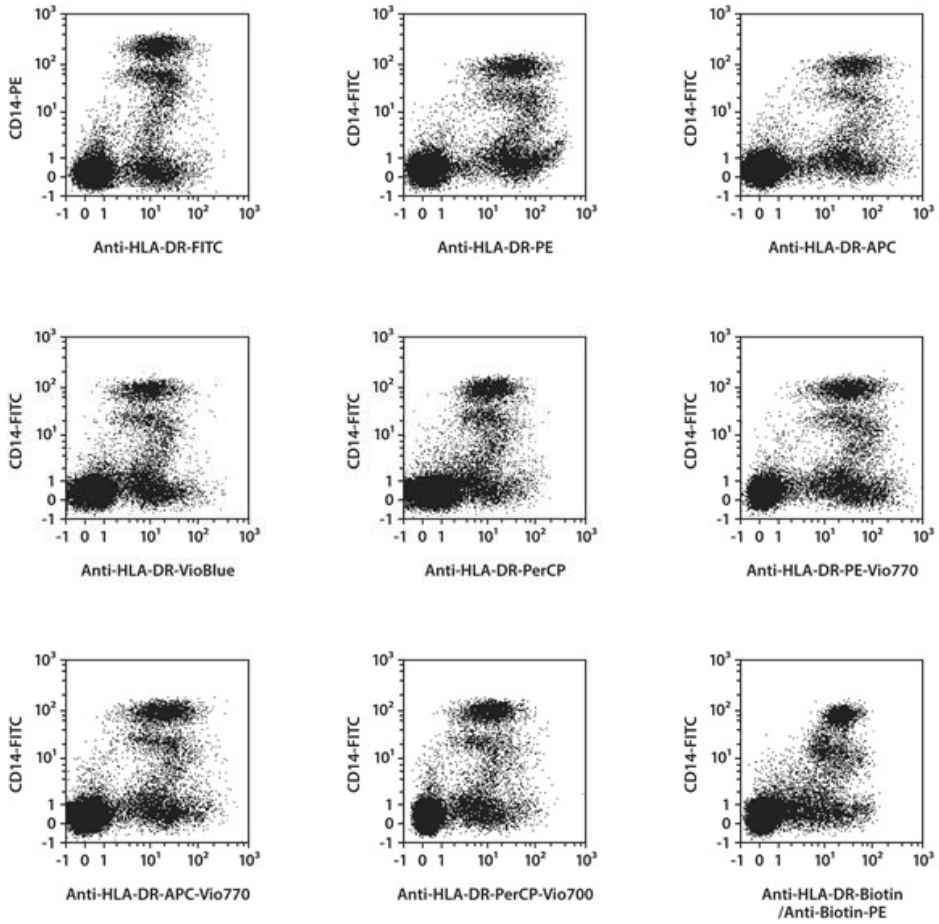
- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:11 for up to 10^7 cells/100 µL of buffer.
- Volumes given below are for up to 10^7 nucleated cells. When working with fewer than 10^7 cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes accordingly (e.g. for 2×10^7 nucleated cells, use twice the volume of all indicated reagent volumes and total volumes).

1. Determine cell number.
2. Centrifuge cell suspension at 300×g for 10 minutes. Aspirate supernatant completely.
3. Resuspend up to 10^7 nucleated cells per 100 µL of buffer.
4. Add 10 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 100 µL of buffer, add 10 µL of fluorochrome-conjugated anti-biotin antibody, and continue as described in steps 5 and 6.
8. Resuspend cell pellet in a suitable amount of buffer for analysis by flow cytometry or fluorescence microscopy.

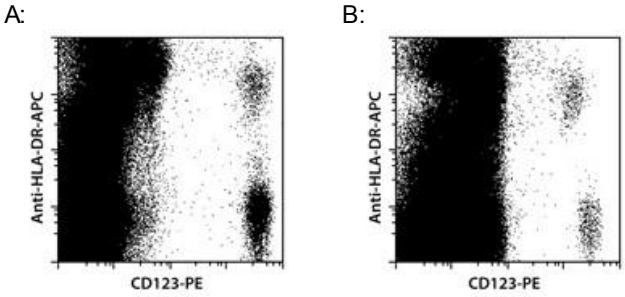
Examples of immunofluorescent staining

Human peripheral blood mononuclear cells (PBMCs) were stained with Anti-HLA-DR antibodies as well as 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.



Rhesus monkey (A) or cynomolgus monkey (B) PBMCs were stained with Anti-HLA-DR-APC as well as with CD123-PE (clone 7G3). Cells were analyzed by flow cytometry using the MACSQuant® Analyzer.



Warranty

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