Anti-TCR Vα2 antibodies, mouse

For research use only

9 µg equal 60 tests, 30 µg equal 200 tests. One test corresponds to labeling of 10^6 cells.

Product	Content	Order no.
Anti-TCR Vα2-FITC	9 µg in 300 µL	130-110-174
Anti-TCR Vα2-FITC	30 µg in 1 mL	130-110-148
Anti-TCR Vα2-PE	9 µg in 300 µL	130-110-175
Anti-TCR Vα2-PE	30 µg in 1 mL	130-110-149
Anti-TCR Vα2-APC	9 µg in 300 µL	130-110-176
Anti-TCR Vα2-APC	30 µg in 1 mL	130-110-150
Anti-TCR Vα2-VioBlue	9 µg in 300 µL	130-110-402
Anti-TCR Vα2-VioBlue	30 µg in 1 mL	130-110-301
Anti-TCR Vα2-PE-Vio770	9 µg in 300 µL	130-110-177
Anti-TCR Vα2-PE-Vio770	30 µg in 1 mL	130-110-151
Anti-TCR Vα2-APC-Vio770	9 µg in 300 µL	130-110-178
Anti-TCR Vα2-APC-Vio770	30 µg in 1 mL	130-110-152
Anti-TCR Vα2-Biotin	9 µg in 300 µL	130-110-173
Anti-TCR Vα2-Biotin	30 µg in 1 mL	130-110-147

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	TCR Vα2
Clone	REA679
Isotype	recombinant human IgG1
Isotype control	REA Control antibodies
Alternative names of antigen	TCR Va2, TCR alpha2
Molecular mass of antigen [kDa]	10
Distribution of antigen	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 REA679 recognizes the mouse Vα2 T cell receptor (TCR Vα2) of mice having the a, b, and c haplotypes. The TCR is responsible for recognizing antigens bound to major histocompatibility complex (MHC) molecules. It is a disulfide-linked membrane-anchored heterodimeric glycoprotein normally consisting of the highly variable α and β chains expressed as part of a complex with the invariant CD3 chain molecules. TCR Vα elements are expressed preferentially in CD4 or CD8 subsets in a manner that is largely independent of the MHC haplotype. It is likely that the Vα elements interact preferentially with conserved regions of class I or class II molecules.
Additional information: Clone REA679 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.

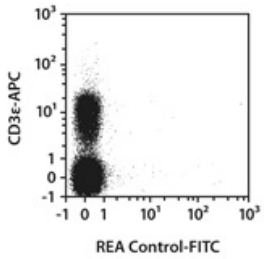
Protocol for cell surface staining

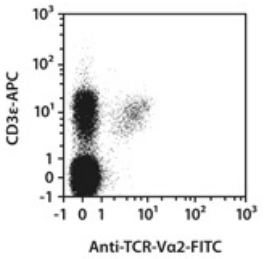
- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:10 for up to 10^6 cells/50 µL of buffer.
- Volumes given below are for up to 10^6 nucleated cells. When working with fewer than 10^6 cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes accordingly (e.g. for 2×10^6 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 10^6 nucleated cells per 45 µL of buffer.
4. Add 5 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

Splenocytes from C57BL/6 mice were stained with Anti-TCR Vα2 antibodies or with the corresponding REA Control antibodies (left image) as well as with CD3ε 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.


CD3ε-APC
REA Control-FITC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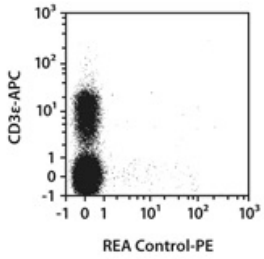
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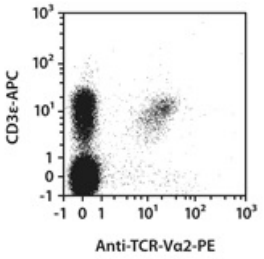
REA Control-PE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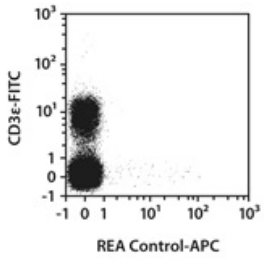
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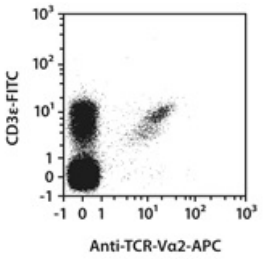
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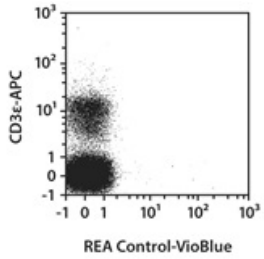
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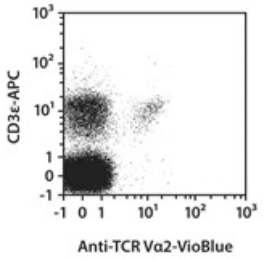
REA Control-VioBlue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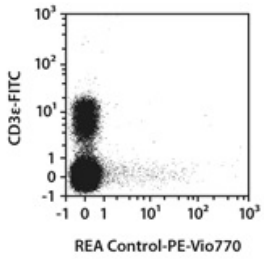
Anti-TCR Vα2-VioBlue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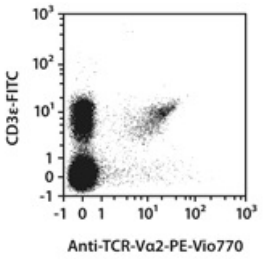
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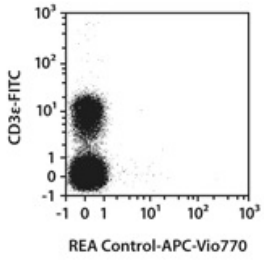
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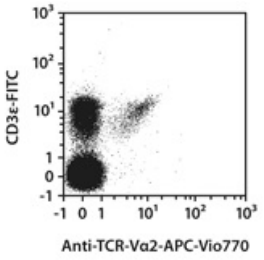
REA Control-APC-Vio770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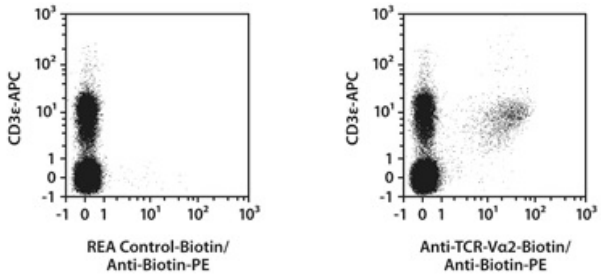
Anti-TCR-Vα2-APC-Vio770



References

1. **Kubo, R. T. *et al.*** (1989) Characterization of a monoclonal antibody which detects all murine alpha beta T cell receptors. J. Immunol. 142(8): 2736–2742.
2. **Grégoire, C. *et al.*** (1991) Engineered secreted T-cell receptor alpha beta heterodimers. Proc. Natl. Acad. Sci. U.S.A. 88(18): 8077–8081.

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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**
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