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Monoclonal Antibody to T-Cell Receptor TCR Vb 8.1, 8.2 - Purified

Alternate names: TCR V beta8, TCR Vb8

Catalog No.: CL081P

Quantity: 0.2 mg

Concentration: 0.2 mg/ml

Host / Isotype: Mouse / IgG2a

Recommended Isotype Controls: AM03096PU-N

Clone: KT-8E

Format:
State: Liquid, purified.
Buffer System: PBS and 0.09% NaN3.

Applications:
Flow cytometry (see protocol).
Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

Specificity:
This monoclonal antibody reacts with mouse TCR Vb 8.2, 8.3, bearing T cells.
Species: Mouse.
Other species not tested.

Storage:
Store the antibody undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer.
Avoid repeated freezing and thawing.
Shelf life: one year from despatch.

General Readings:
1. Tomonari, K., Immunogenetics (in press).

Protocols:

FLOW CYTOMETRY ANALYSIS:

Method:
1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population with Lympholyte®-M cell separation medium.
2. Wash 2 times.
3. Resuspend the cells to a concentration of 2x10e7 cells/ml in media A. Add 50 µl of this suspension to each tube (each tube will then contain 1x10e6 cells, representing 1 test).
4. To each tube, add ~1.0 µg of this Ab.
5. Vortex the tubes to ensure thorough mixing of antibody and cells.
6. Incubate the tubes for 30 minutes at 4°C.
7. Wash 2 times at 4°C.
8. Add 100 µl of secondary antibody (FITC Goat anti-mouse IgG (H+L)) at 1:500 dilution.
9. Incubate the tubes at 4°C for 30-60 minutes. (It is recommended that the tubes are protected from light since most fluorochromes are light sensitive).

10. Wash 2 times at 4°C in media B.
11. Resuspend the cell pellet in 50 µl ice cold media B.
12. Transfer to suitable tubes for flow cytometric analysis containing 15 µl of propidium iodide at 0.5 mg/ml in PBS. This stains dead cells by intercalating in DNA.

Media:
A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide (100 µl of 2M sodium azide in 100 mls).
B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 µl of 2M sodium azide in 100 mls).