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CI 077B

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Monoclonal Antibody to T Cell Receptor (TCR) Vb 4 - Biotin

Alternate names: TCR V beta-4, TCR Vb4

Catalog No.: CL077B

Quantity: 0.1 mg

Concentration: 0.1 mg/ml

Host / Isotype: Rat / IgG2a

Clone: CTVB4

Format: State: Liquid purified

Buffer System: PBS, 0.09% NaN3 and EIA grade BSA as a stabilizing protein to bring total

protein concentration to 4-5 mg/ml.

Label: Biotin

Applications: Flow cytometry: Blocking Fc receptors with an anti-mouse CD16/32 mAb can significantly

reduce background staining by the CTVB4 mAb.

Other applications not tested. Optimal dilutions are dependent on conditions and should

be determined by the user.

Specificity: This monoclonal antibody reacts with TCR Vβ4 bearing T cells. TCR Vβ4 is expressed in

most known mouse strains.

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., et al. 1990. Correlation between the Vβ4+ CD8+ T cell population and the

H-2d haplotype. Immunogenetics 31:333-339.

2. Padula, S.J., et al. 1991. Identification of encephalitogenic Vβ4-bearing T cells in SJL mice-

further evidence for the V region disease hyopothesis J. Immunol. 146:879-883.

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.

Wash 2 times.

3. Resuspend the cells to a concentration of 2x10e7 cells/ml in media A. Add 50 μ l of this suspension to each tube (each tube will then contain 1 x 10e6 cells, representing 1 test).

4. To each tube, add \sim 1.0 μ g* of this Ab per 10e6 cells.

5. Vortex the tubes to ensure thorough mixing of antibody and cells.

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- 6. Incubate the tubes for 30 minutes at 4°C.
- 7. Wash 2 times at 4°C.
- 8. Add 100 µl of (Streptavidin-PE) at a 1:50 dilution.
- 9. Incubate tubes at 4°C for 30 60 minutes (It is recommended that tubes are protected from light since most fluorochromes are light sensitive).
- 10. Wash 2 times at 4°C.
- 11. Resuspend the cell pellet in 50 µl ice cold media B.
- 12. Transfer to suitable tubes for flow cytometric analysis containing 15 μ l 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 μ l of 2M sodium azide in 100 mls).
- B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 μ l of 2M sodium azide in 100 mls).

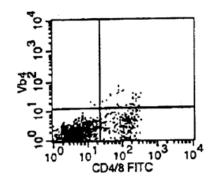
Results - Tissue Distribution by Flow Cytometry Analysis:

(Representative Dot Blot) Mouse Strain: C3H.SW

<u>Cell Concentration</u>: 1x10e6 cells per tests <u>Antibody Concentration Used</u>: 1.0 μg/10e6 cells

Isotypic Control: Biotin Rat IgG2a

Pictures:



Cell source: Spleen
Percentage of cells stained above control: 2.7%

