

CL004B**Monoclonal Antibody to CD4 - Biotin**

Alternate names:	T-cell surface antigen T4/Leu-3, T-cell surface glycoprotein CD4
Quantity:	0.1 mg
Concentration:	0.1 mg/ml
Background:	CD4 (L3T4) which is expressed on the majority of thymocytes and on the MHC class II restricted subset of mature T cells including Th cells ^{1,2} . Mouse CD4 has also been reported to be present on multipotential hematopoietic stem cells, bone marrow myeloid precursors, and intrathymic precursors ^{2,3} . As a coreceptor in the TCR complex, CD4 is involved in T cell activation through interaction with MHC class II on APC's and in signal transduction via protein tyrosine kinase lck1.
Uniprot ID:	P06332
NCBI:	NP_038516.1
GeneID:	12504
Host / Isotype:	Rat / IgG2a
Clone:	CT-CD4
Format:	State: Liquid purified IgG fraction Buffer System: PBS containing 0.09% Sodium Azide and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml Label: Biotin
Applications:	Flow Cytometry (See Protocols). (Reported to be useful in immunohistochemistry on Acetone Fixed Frozen Sections). Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Specificity:	This CT-CD4 monoclonal antibody (mAb) recognizes Mouse CD4 (L3T4). 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. Bierer, B.E. et al. 1989. Annu. Rev. Immunol. 7: 579-599. 2. Fredrickson, G.G. et al. 1989. J. Exp. Med. 169: 1473-1478. 3. Wu, L. et al. 1991. Nature 349: 71-74. Dialynas, D.P. et al 1983. J. Immunol. 131:2445-2451. 4. Palathumpat, V. et al. 1992. J. Immunol. 148:3319-3326. 5. Gross, J. A. et al. 1992. J. Immunol. 149:380-388. 6. Darby, C.R. et al. 1993. J. Immunol. 159:125-129. 7. Darby, C.R. et al. 1992. Transplantation 54:483-490. 8. Cobbold, S.P. et al. 1984. Nature 312:548-551. 9. Agel, N.M. et al 1984. J. Immunol. Methods 69:207-214.

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 2×10^7 cells/ml in media A. Add 50 μ l of this suspension to each tube (each tube will then contain 1×10^6 cells, representing 1 test).
4. To each tube, add ~1.0-0.5 μ g of this Ab per 10^6 cells.
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 μ l of (Streptavidin-PE) at a 1:20 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 μ l 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).

Tissue Distribution by Flow Cytometry Analysis:

Mouse Strain: BALB/c

Cell Concentration: 1×10^6 cells per test

Antibody Concentration Used: 0.5 μ g/ 10^6 cells

Isotypic Control: Biotin Rat IgG2a

Pictures:

Cell Source: Spleen Percentage of cells stained above control: 18.38%

