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Monoclonal Antibody to CD3 - FITC

Alternate names: T-cell surface antigen T3/Leu-4, T-cell surface glycoprotein CD3, T3/Leu-4

Catalog No.: CL002F

Quantity: 0.1 mg

Concentration: 0.1 mg/ml

Background: In the mouse, CD3 is highly expressed by mature T cells and at lower levels by CD4+CD8+ thymocytes2. The ε chain of CD3 is a component of the multimeric T cell receptor complex and is involved with signal transduction

Uniprot ID: P22646

NCBI: NP_031674.1

GeneID: 12501

Host / Isotype: Hamster / IgG

Clone: 500-A2

Immunogen: C6VL-BS cell lysate

Format:
State: Liquid purified IgG
Buffer System: PBS, 0.1% NaN3 and EIA grade BSA as stabilizing protien to bring total protein concentration to 4-5 mg/ml
Label: FITC

Applications:
Flow Cytometry (see Protocols).
(Reportet to be useful in immunoprecipitation,
immunofluorescent staining and 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 anti-mouse CD3ε monoclonal antibody is specific for the ε chain of mouse CD31. Immobolized 500A2 mAb can be used to stimulate mouse T cells in vitro1. A combination of immobolized 500A2 mAb and anti-mouse CD28 mAb is commonly used for the activation of mouse T cells3.
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.

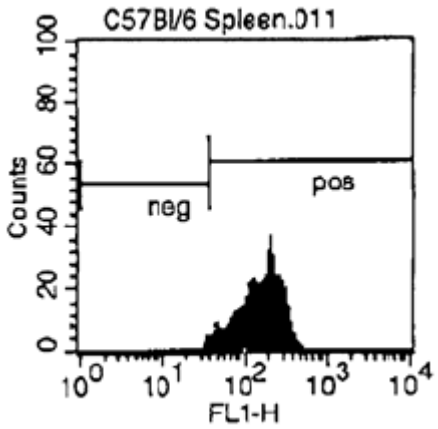
Protocols:
FLOW CYTOMETRY ANALYSIS:
1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population with Lympholyte®-M cell separation medium.
2. Wash 2 times.

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 1 x 10e6 cells, representing 1 test).
4. To each tube, add 0.5 µg* of this Ab per 10e6 cells.
5. Vortex the tubes to ensure thorough mixing of antibody and cells.
6. Incubate the tubes for 30 minutes at 4°C. (It is recommended that the tubes are protected from light, since most fluorochromes are light sensitive.)
7. Wash 2 times at 4°C.
8. Resuspend the cell pellet in 50 µl ice cold media B.
9. 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).

Tissue Distribution by Flow Cytometry Analysis:
Mouse Strain: C57Bl/6
Cell Concentration: 1x10e6 cells per test
Antibody Concentration Used: 0.5 µg/10e6 cells
Isotypic Control: FITC Hamster IgG

Pictures:



Cell Source: Spleen Percentage of cells stained above control: 25.5 %