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Place your order with CEDARLANE[®] or your local distributor. Please contact CEDARLANE[®] for lot specific information.

FITC Anti-Rat CD3 Monoclonal Antibody

CL020F LOT: 21011008

DESCRIPTION:

Cedarlane's anti-rat CD3 monoclonal antibody recognizes a rat T cell surface antigen having a molecular weight of 25,000 Da. This surface antigen appears to be associated with a 92,000 Da heterodimer with subunits of 52,000 and 43,000 Da.

The antibody reacts with rat T cells but not B cells. Immunohistochemically it stains the lymphocytes in the periarterial lymphatic sheaths of the spleen. It stains the medullary cells of the thymus very strongly and the cortical cells much more weakly.

The antibody has the ability to induce rat T cell proliferation in the presence of PMA or when cross - linked to a solid support.

All of these properties resemble those of CD3 in human and mouse systems (1).

This antibody is suitable for use in flow cytometry.

PRESENTATION:

100 µg FITC conjugated Ig buffered in PBS, pH 7.2, and containing 0.09% NaN₃ and 0.5% (w/v) BSA.

STORAGE/STABILITY:

Stable at 4°C. If the reagent is being diluted, it is recommended that only the quantity to be used within one week be diluted. Avoid prolonged exposure to light. **SPECIFICATIONS:**

Clone: 1F4

Specificity: Rat CD3

Ig Class: Mouse IgM

Antibody Concentration: 0.5 mg/ml

EDARLA

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Continued Overleaf.....

For more information or to place an order please contact...

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FLOW CYTOMETRY ANALYSIS:

Method:

- 1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population with Lympholyte[®]-Rat cell separation medium (CL5040).
- 2. Wash 2 times.
- 3. Resuspend the cells to a concentration of $2x10^7$ cells/ml in media A. Add 50 µl of this suspension to each tube (each tube will then contain 1×10^6 cells, representing 1 test).
- 4. To each tube, add $1.0 \ \mu g^*$ of **CL020F** per 10^6 cells.
- 5. Vortex the tubes to ensure thorough mixing of antibody and cells.
- 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 μ l 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).

Results:

<u>Tissue Distribution by Flow Cytometry Analysis</u>: **Representative Histogram**

Cell Concentration : $1x10^{6}$ cells per test Antibody Concentration Used: $1.0 \mu g/10^{6}$ cells Isotypic Control: FITC Mouse IgM (CLCMGM01)



Cell Source: Spleen

N.B. Appropriate control samples should always be included in any labelling studies.

* For optimal results in various applications, it is recommended that each investigator determine dilutions appropriate

<u>REFERENCES</u>:

- 1. Nicolls, M.G., Aversa, G.A., Pearce, N.W., Spinelli, A., Berger, M.F., Gurley, K.E. and Hall, B.M. 1992 Induction of long term specific tolerance to allografts in vivo by therapy with a monoclonal antibody to rat CD3. Transplantation 55:459-468.
- 2. Tanaka, T., Masuko, T., Yagita, H., Tamura, T. and Hashimota, Y. 1989 Characterization of a CD3-like rat T cell surface antigen recognized by a monoclonal antibody. J. Immunol. 142:2791-2795.

Laboratory Reagent For Research Use Only

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JV 10/02/08