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TECHNICALLY Speaking

Place your order with CEDARLANE® or your local distributor.

Please contact CEDARLANE® for lot specific information.

FITC Anti-Rat CD25 (IL-2R) Monoclonal Antibody

CL039F
CL039F-5
LOT: 3932

DESCRIPTION:

Cedarlane's anti-rat Interleukin-2 receptor monoclonal antibody recognizes the smaller (alpha subunit) 55kD chain of the IL-2 receptor found on activated rat T cells, thymic dendritic cells but not resting lymphocytes (1). CL039F binds to the rat interleukin-2 receptor and has proven to be an important marker for activated T cells. (2).

This clone is reported to work with frozen and paraffin sections (3).

PRESENTATION:

100 µg (**CL039F**) or 500 µg (**CL039F-5**) FITC conjugated Ig buffered in PBS, 0.02% NaN_3 and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml.

STORAGE/STABILITY:

Store at 4°C. For long term storage, aliquot and freeze unused portion at -20°C in volumes appropriate for single usage. Avoid freeze/thaw cycles. Avoid prolonged exposure to light.

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

CEDARLANE® LABORATORIES LIMITED

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or visit our website for a list of our international distributors including contact information

website: www.cedarlanelabs.com • e-mail: info@cedarlanelabs.com

SPECIFICATIONS:

Clone: MRC OX-39

Hybridoma Production:

Immunization: Immunogen: T blasts from a mixed lymphocyte reaction between purified CD4 positive T cells and irradiated spleens.
Donor: BALB/c spleen

Fusion Partner: NSO/1

Specificity: Rat CD25 (IL-2R)

Ig Class: Mouse IgG_1

Format: FITC conjugated Ig buffered in PBS, 0.02% NaN_3 and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml. (Purified from ascitic fluid via Protein G Chromatography)

Antibody Concentration: 0.1 mg/ml

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 0.05-0.1 µg***** of **CL039F or CL039F-5** 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.
   (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:

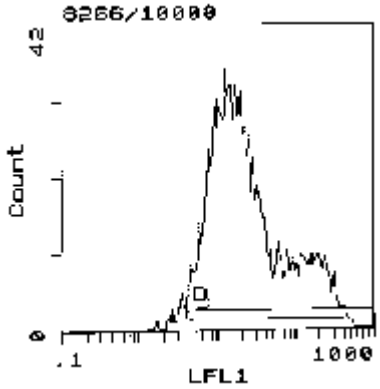
A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide (100 µl of 2M sodium azide in 100 mls).
B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 µl of 2M sodium azide in 100 mls).

Results:

Tissue Distribution by Flow Cytometry Analysis:

Rat Strain: Wistar
Cell Concentration : $1x10^6$ cells per test
Antibody Concentration Used: 0.1 µg/10^6 cells
Isotypic Control: FITC Mouse IgG_1(CLCMG101)

Cell Source	Percentage of cells stained above control:
T Cell Blasts	97.5%
Thymus	11.8%



Cell Source: T Cell blasts
Percentage of cells stained above control: 97.5%

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 for individual use.**

REFERENCES:

1) Paterson, D.J., Jeffries, W.A., Green, J.R., Brandon, M.R., Corthesy, P., Puklavec, M. and A.F. Williams. (1987) Mol. Immunol. 24, 1281-1290. Antigens of Activated Rat T Lymphocytes Including a Molecule of 50,000 Mr Detected Only on CD4 Positive T Blasts

2) Barclay, A.N. (1981) Immunology. 42 593-600 The Localization of populations of lymphocytes defined with monoclonal antibodies in rat lymphoid tissues.

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