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Conveniently Delivering You Today's Innovations
for the Science of Tomorrow™

**Purified Anti-Rat CD25 (IL-2R)
Monoclonal Antibody**

CL039AP
CL039AP-2
LOT: 1039218A

DESCRIPTION:

Cedarlane's anti-rat CD25 (IL-2R) 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). CL039AP binds to the rat interleukin-2 receptor designated CD25 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:

250 µg (**CL039AP**) or 500 µg (**CL039AP-2**) purified Ig buffered in PBS and 0.02% NaN₃.
For maximal recovery of contents, please quick-spin vial before opening.

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.

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-2 Receptor)

Ig Class: Mouse IgG₁

Format: Purified Ig buffered in PBS and 0.02% NaN₃. (Purified from ascitic fluid via Protein G Chromatography)

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CEDARLANE[®] 

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An ISO 9001:2000 and ISO 13485:2003
registered company.

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Antibody Concentration: 1.0 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 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 μ g* of **CL039AP or CL039AP-2**.
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 secondary antibody **CLCC30204** (PE Goat anti-mouse IgG (H+L)) at 1:20 dilution.
9. Incubate the tubes at 4°C for 30-60 minutes.
(It is recommended that the tubes are protected from light since most fluorochromes are light sensitive).
10. Wash 2 times at 4°C in media B.
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).

Results:

Tissue Distribution by Flow Cytometry Analysis:

Rat Strain: Wistar

Cell Concentration : 1×10^6 cells per test

Antibody Concentration Used: 1.0 μ g/ 10^6 cells

Isotypic Control: Mouse IgG₁, kappa

Cell Source

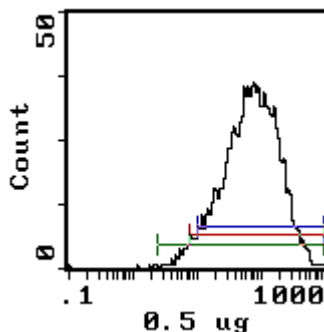
Percentage of cells stained above control:

Thymus - unactivated

2.51%

Con-A activated thymocytes

93.98%



LFL2

Cell Source: Con-A Activated Thymocytes

Percentage of cells stained above control: 93.98%

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.
- 3) Whiteland, J.L et al (1995). Immunohistochemical detection of T cell subsets and other leukocytes in paraffin embedded rat and mouse tissues with monoclonal antibodies .J. Histochem. Cytochem. 43: 313-320.

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