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### SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

[mail@szabo-scandic.com](mailto:mail@szabo-scandic.com)

[www.szabo-scandic.com](http://www.szabo-scandic.com)

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# TECHNICALLY *Speaking*

Place your order with CEDARLANE® or your local distributor.

*Please contact CEDARLANE® for lot specific information.*

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

**CL039PE  
CL039PE-4  
LOT: 3951**

### **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). CL039PE 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:**

50 µg (CL039PE) or 200 µg (CL039PE-4) PE conjugated Ig buffered in PBS, 0.02% NaN<sub>3</sub> and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml.

### **STORAGE/STABILITY:**

Store at 4°C. **DO NOT FREEZE.** Avoid prolonged exposure to light.

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

**CEDARLANE®**  
**LABORATORIES LIMITED**



*toll free: 1-800-268-5058  
in North America*

phone: (905) 878-8891 • fax: (905) 878-7800

5516 - 8th Line, R.R.#2, Hornby, Ontario, CANADA L0P 1E0

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

website: [www.cedarlanelabs.com](http://www.cedarlanelabs.com) • e-mail: [info@cedarlanelabs.com](mailto: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<sub>1</sub>

Format: R-PE conjugated Ig buffered in PBS, 0.02% NaN<sub>3</sub> 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<sup>®</sup>-Rat Cell Separation Medium (CL5040).
2. Wash 2 times.
3. Resuspend the cells to a concentration of  $2 \times 10^7$  cells/ml in media A. Add 50  $\mu$ l of this suspension to each tube (each tube will then contain  $1 \times 10^6$  cells, representing 1 test).
4. To each tube, add 0.2-0.5  $\mu$ g\* of **CL039PE or CL039PE-4** 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 fluorochemicals are light sensitive.)
7. Wash 2 times at 4°C.
8. Resuspend the cell pellet in 50  $\mu$ l ice cold media B.
9. Transfer to suitable tubes for flow cytometric analysis containing 15  $\mu$ 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 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: Buffalo

Cell Concentration :  $1 \times 10^6$  cells per test

Antibody Concentration Used:  $0.2 \mu\text{g}/10^6$  cells

Isotypic Control: PE Mouse IgG<sub>1</sub>

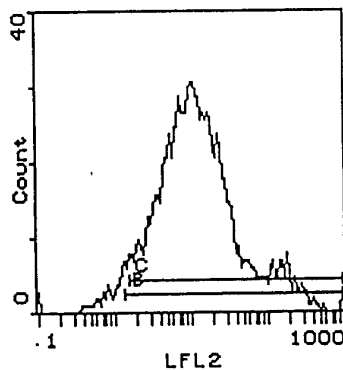
Cell SourcePercentage of cells stained above control:

T Cell Blasts

95.3%

Thymus

13.3%



Cell Source: T Cell blasts

Percentage of cells stained above control: 95.3%

**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.**

R-Phycoerythrin conjugates are produced under license and protected under Stanford University held patents 4,520,110; 4,542,104; 4,859,582; 5,055,556 (U.S.); 76695 (EPC); 548440 (Australia); 1,179,942 (Canada); and 1,594,827 (Japan).

**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. Antigen 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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