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TECHNICALLY *Speaking*

Place your order with CEDARLANE® or your local distributor.

Please contact CEDARLANE® for lot specific information.

Biotin Anti-Rat $\alpha\beta$ TCR Monoclonal Antibody

CL057B
CL057B-5
LOT: 5741

DESCRIPTION:

Cedarlane's anti-rat $\alpha\beta$ T cell receptor monoclonal antibody detects a T cell-specific heterodimeric 40 and 46 kD cell surface protein that is expressed on the vast majority, but not all, peripheral rat T cells. CL057B induces blastoid transformation and IL-2R expression in T-cells when cross linked on an artificial surface. It also induces cell proliferation when exogenous IL-2 is added.

Applications: Flow cytometry, immunohistochemical staining on frozen sections.

PRESENTATION:

100 μg (**CL057B**) or 500 μg (**CL057B-5**) Biotin 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.

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

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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: R73

Hybridoma Production:

Immunization: Immunogen: Rat T Blasts

Donor: BALB/c spleen

Fusion Partner: X63-Ag 8.653

Specificity: Rat $\alpha\beta$ TCR

Ig Class: Mouse IgG₁

Format: Biotin conjugated Ig buffered in PBS, 0.02% NaN₃ and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml. (Purified from Bioreactor supernatant 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[®]-R 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 0.2-0.5 μ g* of **CL057B** or **CL057B-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.
7. Wash 2 times at 4°C.
8. Add 100 μ l of secondary antibody **CLCSA1001** (Streptavidin-FITC) at a 1:500 dilution.
9. Incubate tubes at 4°C for 30 - 60 minutes (It is recommended that tubes are protected from light since most fluorochromes are light sensitive).
10. Wash 2 times at 4°C.
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 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 : 1×10^6 cells per test

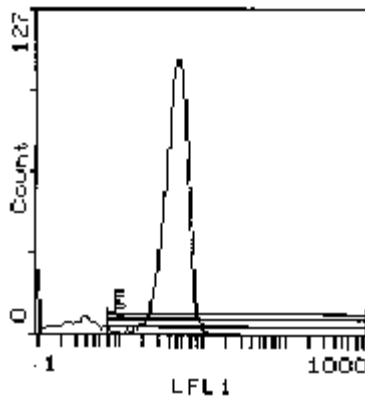
Antibody Concentration Used: $0.2 \mu\text{g}/10^6$ cells

Isotypic Control: Biotin Mouse IgG₁(CLCMG115)

Cell SourcePercentage of cells stained above control:

Thymus	66.3%
Splenic T Cells*	89.9%

*(T cells isolated with CL102 - Cedarlane's Rat T Cell Recovery Column Kit)



Cell Source: Splenic T Cells

Percentage of cells stained above control: 89.9%

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.**

Strain Distribution by Flow Cytometry Analysis:

Procedure: see page 2

Cell Concentration : 1×10^6 cells per test

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

Strains Tested: Buffalo, Wistar, Fischer 344, Brown Norway

Positive: Buffalo, Wistar, Fischer 344, Brown Norway

Negative: none

REFERENCES:

1. Hunig, T., Wallny, H.J., Hartley, J.K., Lawetzky, A., and Tiefenthaler, G. (1989). A Monoclonal Antibody to a Constant Determinant of the Rat T-Cell Antigen Receptor that Induces T-Cell Activation. *J. Exp. Med.* 169:73-86.

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