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TECHNICALLY Speaking

Place your order with CEDARLANE® or your local distributor.

Please contact CEDARLANE® for lot specific information.

Purified Anti-Rat γδ TCR Monoclonal Antibody

CL056AP
CL056AP-2
LOT: 5621

DESCRIPTION:

Cedarlane's anti-rat γδ T cell receptor monoclonal antibody detects a T cell-specific heterodimeric 48 and 50 kD cell surface protein that is expressed on greater than 90% of $CD3^+$ αβ TCR– rat peripheral T lymphocytes, and identifies a dense network of dendritic cells in the epidermis as γ/δ T cells. Immobilized this antibody induces a strong proliferative response in γδ T cell cultures supplemented with either IL-2 or IL-4.

Applications: immunoprecipitation, flow cytometry, immunohistochemical staining on frozen sections.

PRESENTATION:

250 µg (CL056AP) or 500 µg (CL056AP-2) purified Ig buffered in PBS and 0.02% NaN_3.

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 repeated freeze/thaw cycles.

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: V65

Hybridoma Production:

Immunization: Immunogen: Rat T Blasts
Donor: BALB/c Spleen

Fusion Partner: X63-Ag 8.653

Specificity: Rat γδ TCR

Ig Class: Mouse IgG_1

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 $2x10^7$ cells/ml in media A. Add 50 µl of this suspension to each tube (each tube will then contain $1x10^6$ cells, representing 1 test).
4. To each tube, add 0.5-1.0 µg***** of **CL056AP or CL056AP-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 µl 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 µl ice cold media B.
12. 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).

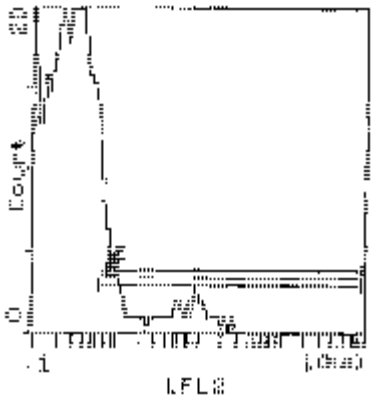
Results:

Tissue Distribution by Flow Cytometry Analysis:

Rat Strain: Fischer
Cell Concentration : $1x10^6$ cells per test
Antibody Concentration Used: 0.5 µg/10^6 cells
Isotypic Control: Mouse IgG_1

Cell Source	Percentage of cells stained above control:
Thymus	4.2%
Splenic T Cells*	5.6%

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



Cell Source: Splenic T-cells
Percentage of cells stained above control: 5.6%

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. Kühnlein, P., Park, H-J., Herrmann, T., Elbe, A., and Hünig, T. (1994). Identification and Characterization of Rat γδ T-Lymphocytes in Peripheral Lymphoid Organs, Small Intestine, and Skin With a Monoclonal Antibody to a Constant Determinant of the γ/δ T cell Receptor. J. Immunol. 153:979-986.

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