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TECHNICALLY Speaking

Place your order with CEDARLANE® or your local distributor.

Please contact CEDARLANE® for lot specific information.

Biotin Anti-Mouse CD8a Antigen Monoclonal Antibody

CL168B
CL168B-3
LOT: 12020708

DESCRIPTION:

Cedarlane's anti-mouse CD8a antigen monoclonal antibody recognizes the mouse CD8α chain. The α chain of CD8 associates with the CD8β chain to form a CD8α/βheterodimer that is expressed by the majority of thymocytes and by the MHC class I restricted subset of mature T cells[1]. Mouse CD8α can also form a CD8α/α chain homodimer on subsets of CD8 positive T cells. For this reason antibodies specific for CD8a rather than CD8b are recommended for a rigorous delineation of CD8 positive cells.

This antibody is suitable for use in flow cytometry.

PRESENTATION:

100 µg (**CL168B**) or 300 µg (**CL168B-3**) Biotin conjugated Ig buffered in PBS, 0.1% sodium azide (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. If the reagent is being diluted, it is recommended that only the quantity to be used within one week be diluted. Check label for expiry date.

SPECIFICATIONS:

Clone: CT-CD8a

Specificity: Mouse CD8a (Ly-2)

Ig Class: Rat IgG_{2a}

Antibody Concentration: 0.1 mg/ml

Continued Overleaf….

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

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FLOW CYTOMETRY ANALYSIS:

Method:

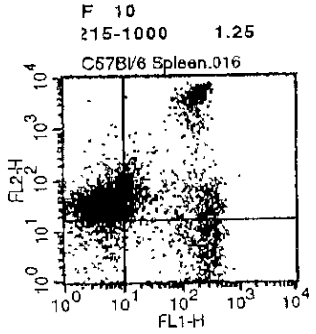
1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population with Lympholyte®-M cell separation medium (CL5030).
2. Wash 2 times.
3. Resuspend the cells to a concentration of $2x10^7$ cells/ml in media A. Add 50 µl of this suspension to each tube (each tube will then contain 1×10^6 cells, representing 1 test).
4. To each tube, add ~0.125 µg* of **CL168B or CL168B-3** 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 µl of secondary antibody **CLCSA1004** (Streptavidin-PE) at a 1:5 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 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:
Representative Dot Plot
Mouse Strain: BALB/c
Cell Concentration: $1x10^6$ cells per test
Antibody Concentration Used: 0.125 µg/10^6 cells
Isotypic Control: Biotin Rat IgG_{2a} (CLCR2A15)



Cell Source: Spleen

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. Tomonari, K. and Spencer, S. 1990 Epitope-specific binding of CD8 regulates activation of T cells and induction of cytotoxicity. International Immunology 2(12):1189-1194.

FOR RESEARCH USE ONLY

JV 06/26/07