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TECHNICALLY Speaking

Place your order with CEDARLANE® or your local distributor.

Please contact CEDARLANE® for lot specific information.

FITC Anti-Mouse I-A^b Monoclonal Antibody

CL8702F
LOT:

DESCRIPTION:

Cedarlane's anti-mouse I-A^b monoclonal antibody reacts with the I-A^b encoded MHC class II antigen expressed on mouse strains of the H-2^b haplotype. It also reacts with the I-A^d encoded MHC class II antigen expressed on mouse strains of the H-2^d haplotype. Class II antigens are most highly expressed on antigen-presenting cells including B cells, macrophages, dendritic cells and certain epithelial cells.

PRESENTATION:

100 µg FITC 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 and prolonged exposure to light. 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.

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

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website: www.cedarlanelabs.com • e-mail: info@cedarlanelabs.com

SPECIFICATIONS:

Clone: 28-16-8S

Specificity: Mouse MHC class II I-A^b, cross reacts with I-A^d

Ig Class: Mouse IgM

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®-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 $1x10^6$ cells, representing 1 test).
4. To each tube, add~1.0 µg of **CL8702F** 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 fluorochromes are light sensitive).
7. Wash 2 times at 4°C.
8. Resuspend the cell pellet in 50 µl ice cold media B.
9. 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.

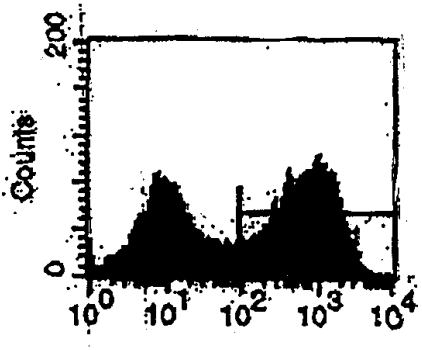
Media:

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:

Mouse strain: C57BL/6
Cell Concentration: $1x10^6$ cells per test
Antibody Concentration Used: 1.0 µg/10^6 cells
Isotypic Control: FITC Mouse IgM (CLCMGM01)



LFL 1
Cell Source: Spleen
Percentage of cells stained above control: 56.7%

N.B. Appropriate control samples should always be included in labelling studies.

*** For optimal results in various applications, it is recommended that each investigator determine dilutions appropriate for individual use.**

REFERENCES:

1. Ozato, K., and Sachs, D.H. 1981. Monoclonal antibodies to mouse MHC Antigens. III. Hybridoma antibodies reacting to antigens of the H-2^b haplotype reveal genetic control of isotype expression. J. Immunol. 126:317-322.

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