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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 CD62L Monoclonal Antibody

CL8918F
CL8918F-3
LOT: 1831

DESCRIPTION:

Cedarlane's anti-mouse CD62L (L-selectin Ly 22) monoclonal antibody reacts with a 90 kDa protein which is involved with the homing of lymphocytes to peripheral lymph nodes.

L-selectin is expressed on most T and B lymphocytes, neutrophils, monocytes, eosinophils.[1] Pre-incubation of lymphocytes with this antibody completely and specifically blocks binding of lymphocytes to high endothelial venules (HEV) in vitro [2,3,6] and the migration of lymphocytes to lymph nodes in vivo.[2,3] Polymorphonuclear cells preincubated with this antibody do not migrate to the inflammatory foci.[3]

Applications of CL8918F include flow cytometry[3,4,5] and IHC.[4]

PRESENTATION:

100 µg (CL8918F) or 300 µg (CL8918F-3) FITC 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. Avoid prolonged exposure to light.

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

CEDARLANE® LABORATORIES LIMITED

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in North America

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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: MEL-14

Hybridoma Production:

Immunization: Immunogen: Mouse B cell Lymphoma, 38C-14
Donor: Fischer Rat Spleen
Fusion Partner: P3 X 63Ag8.653

Specificity: Mouse CD62L (L-selectin Ly 22)

Ig Class: Rat IgG_{2a}

Format: FITC 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. (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®-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 0.2 - 0.5 µg***** of **CL8918F or CL8918F-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.
   (It is recommended that the tubes are protected from light, since most flurochromes 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:

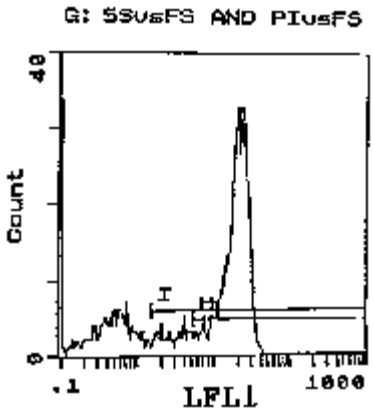
A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide (100 µl of 2M sodium azide in 100 mls).
B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 µl of 2M sodium azide in 100 mls).

Results:

Tissue Distribution by Flow Cytometry Analysis:

Mouse Strain: BALB/c
Cell Concentration : $1x10^6$ cells per test
Antibody Concentration Used: 0.2 µg/10^6 cells
Isotypic Control: FITC Rat IgG_{2a}

Cell Source	Percentage of cells stained above control:
Thymus	88.8%
Spleen	36.6%
Lymph Node	74.4%



Cell Source: Lymph Node
Percentage of cells stained above control: 74.4%

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: $1x10^6$ cells per test
Antibody Concentration Used: 0.2 µg/10^6 cells
Strains Tested: BALB/c, CBA/J, C3H/He, C57BL/6, AKR
Positive: BALB/c, CBA/J, C3H/He, C57BL/6, AKR
Negative: none

REFERENCES:

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2) Gallatin,W.M., I.L. Weissman., E.C. Butcher 1983. A cell surface molecule involved in organ specific homing of lymphocytes. Nature 304:30-34
3) Lewinsohn, D.M., R.F. Bargatze, E.C. Butcher 1987. Leukocyte-endothelial cell recognition: evidence of a common molecular mechanism shared by neutrophils, lymphocytes and other leukocytes. J.Immunology 138:4313-4321
4) Reichert, R., M. Gallitin, E. Butcher, *et al*.. 1984. A homing receptor-bearing cortical thymocyte subset: Implications for thymus cell migration and the nature of cortisone-resisatant thymocytes. Cell 38: 89-99
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