

## Monoclonal Antibody to CD62L / L-Selectin - PE

<b>Alternate names:</b>	CD62 antigen-like family member L, LAM-1, LECAM1, LNHR, LYAM1, Leu-8, Leukocyte adhesion molecule 1, Leukocyte surface antigen Leu-8, Leukocyte-endothelial cell adhesion molecule 1, Lymph node homing receptor, SELL, TQ1, gp90-MEL
<b>Catalog No.:</b>	CL032R
<b>Quantity:</b>	50 µg
<b>Concentration:</b>	0.1 mg/ml
<b>Background:</b>	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.
<b>Uniprot ID:</b>	<a href="#">P18337</a>
<b>NCBI:</b>	<a href="#">NP_035476.1</a>
<b>GeneID:</b>	<a href="#">20343</a>
<b>Host / Isotype:</b>	Rat / IgG2a
<b>Clone:</b>	MEL-14
<b>Immunogen:</b>	Mouse B cell Lymphoma, 38C-14
<b>Format:</b>	<b>State:</b> Liquid purified Ig fraction <b>Purification:</b> Protein G Chromatography <b>Buffer System:</b> PBS containing 0.02% Sodium Azide as preservative and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml <b>Label:</b> PE
<b>Applications:</b>	<b>Flow Cytometry.</b> Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
<b>Specificity:</b>	This monoclonal antibody reacts with a 90 kDa protein which is involved with the homing of lymphocytes to peripheral lymph nodes.
<b>Species Reactivity:</b>	<b>Tested:</b> Mouse.
<b>Storage:</b>	Store the antibody undiluted at 2-8°C. <b>DO NOT FREEZE!</b> This product is photosensitive and should be protected from light. Shelf life: one year from despatch.
<b>General References:</b>	1) Fink, P., W. Gallatin, R. Reichert, et al. 1985. Homing receptor-bearing thymocytes, an immunocomponent cortical subpopulation. Nature 313: 233-235 2) Gallatin, W.M., I.L. Weissman., E.C. Butcher 1983. A cell surface molecule involved in

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- 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-resistant thymocytes. Cell 38: 89-99
- 5) Siegelman, M., I.C. Cheng, I.L. Weissman, et al. 1990. The mouse lymph node homing receptor is identical with the lymphocyte cell surface receptor Ly-22: Role of the EGF domain in endothelial binding. Cell 61: 611-622
- 6) Jalkanen, S., R.F. Bargatze, J. Toyos, et al. 1987. Lymphocyte Recognition of High Endothelium: Antibodies to Distinct Epitopes of an 85-95-kD Glycoprotein Antigen Differentially Inhibit Lymphocyte Binding to Lymph Node, Mucosal, or Synovial Endothelial Cell. J. of Cell Biol. 105: 983-990
- 7) Goler M.L et al. 1997. T Cell Genetic Background Determines Maintenance of IL-12 Signaling. J. of Immunol. 159: 1767-1774

**Protocols:****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.
2. Wash 2 times.
3. Resuspend the cells to a concentration of  $2 \times 10^7$  cells/ml in media A. Add 50  $\mu$ l of this suspension to each tube (each tube will then contain  $1 \times 10^6$  cells, representing 1 test).
4. To each tube, add 0.5-1.0  $\mu$ g\* of this Ab 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  $\mu$ l ice cold media B.
9. Transfer to suitable tubes for flow cytometric analysis containing 15  $\mu$ 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  $\mu$ l of 2M sodium azide in 100 mls).
- B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100  $\mu$ l of 2M sodium azide in 100 mls).

**Results - Tissue Distribution:**

Mouse Strain: BALB/c

Cell Concentration:  $1 \times 10^6$  cells per test

Antibody Concentration Used: 1.0  $\mu$ g/ $10^6$  cells

Isotypic Control: PE Rat IgG2a

**Cell Source: Percentage of cells stained above control:**

- Thymus: 89.1%  
Spleen: 34.8%  
Lymph Node: 88.5%

**Results - Strain Distribution:**

Cell Concentration:  $1 \times 10^6$  cells per test

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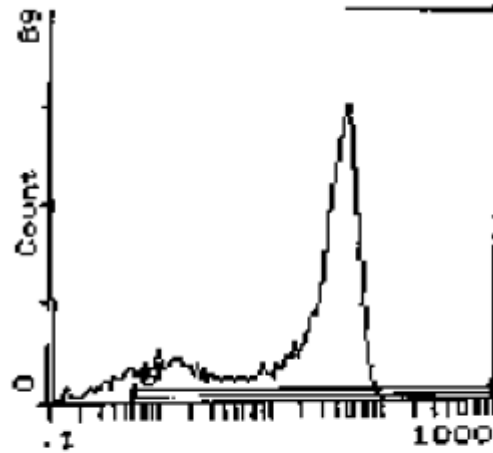
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Antibody Concentration Used: 1.0 µg/10e6 cells  
Strains Tested: BALB/c, CBA/J, C3H/He, C57BL/6, AKR  
Positive: BALB/c, CBA/J, C3H/He, C57BL/6, AKR  
Negative: none

Pictures:



Cell Source: Lymph Node  
Percentage of cells stained above control: 88.5%

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