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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 CD11b Monoclonal Antibody

CL8941F
CL8941F-3
LOT: 4131

DESCRIPTION:

Cedarlane's anti-mouse CD11b (Mac-1; Ly 40) monoclonal antibody is specific for the 170 kDa α subunit of Mac-1 which mediates adhesion to ICAM-1 (CD54) and C3bi. Mac-1 is expressed on granulocytes, macrophages, natural killer cells, and B-1 cells in the peritoneal and pleural cavities. Mac-1 is up-regulated on neutrophils after activation. This particular clone blocks cell adherence and C3bi binding but does not block cell mediated lysis (1,2,3,4,5,6).

Applications include flow cytometry, *in vitro* and *in vivo* blocking, immunohistochemistry (acetone-fixed frozen sections (1-20 μ g/ml), immunoprecipitation and western blotting).

PRESENTATION:

100 μ g (CL8941F) or 300 μ g (CL8941F-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. Check label for expiry date. Avoid prolonged exposure to light.

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

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SPECIFICATIONS:

Clone: M1/70.15

Hybridoma Production:

Immunization: Immunogen: C57BL/10 spleen cell enriched for
T lymphocytes
Donor: DA rat spleen

Fusion Partner: NS-1

Specificity: Mouse CD11b (Mac-1; Ly 40)

Ig Class: Rat IgG_{2b}

Format: FITC conjugated Ig buffered in PBS, 0.02% NaN₃ 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 2×10^7 cells/ml in media A. Add 50 μ l of this suspension to each tube (each tube will then contain 1×10^6 cells, representing 1 test).
4. To each tube, add ~ 0.5 - $1.0 \mu\text{g}^*$ of **CL8941F** or **CL8941F-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 fluorochromes are light sensitive.)
7. Wash 2 times at 4°C .
8. Resuspend the cell pellet in 50 μ l ice cold media B.
9. Transfer to suitable tubes for flow cytometric analysis containing 15 μ 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 μ l of 2M sodium azide in 100 mls).
- B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 μ l of 2M sodium azide in 100 mls).

Results:Tissue Distribution by Flow Cytometry Analysis:

Mouse Strain: BALB/c

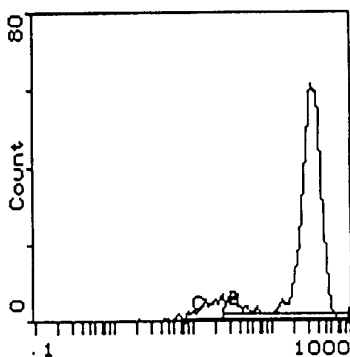
Cell Concentration : 1×10^6 cells per test

Antibody Concentration Used: $0.5 \mu\text{g}/10^6$ cells

Isotypic Control: FITC Rat IgG_{2b}

Cell SourcePercentage of cells stained above control:

Bone Marrow Macrophages	73.1%
Peritoneal Macrophages	89.9%
Thymus	0.7%



LFL1

Cell Source: Peritoneal Macrophages

Percentage of cells stained above control: 89.9%

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:

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2. Springer, T., G. Galfre, D.S. Secher, et al. 1978. Monoclonal xenogeneic antibodies to murine cell surface antigens: identification of novel leukocyte differentiation antigens. *Eur. J. Immunol.* 8:539 - 551.
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