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# TECHNICALLY *Speaking*

Place your order with CEDARLANE® or your local distributor.

*Please contact CEDARLANE® for lot specific information.*

## **PE Rat Anti-Mouse Ly-6A/E (Sca-1) Monoclonal Antibody**

**CL8948PE**  
**CL8948PE-3**  
**LOT: 01010204**

### **DESCRIPTION:**

This monoclonal antibody reacts with Ly-6A.2 and Ly-6E.1, members of the Ly-6 multigene family. Ly-6A/E molecules are expressed on multipotent hematopoietic stem cells<sup>1</sup> activated T cells, and peripheral B cells on mice that express the Ly-6.2 haplotype.

This antibody can be used in immunostaining for flow cytometry<sup>1,4,5</sup>, Western Blot analysis<sup>3</sup>, and immunoprecipitation<sup>1,2</sup>.

### **PRESENTATION:**

50 µg (CL8948PE) or 300 µg (CL8948PE-3) PE conjugated Ig buffered in PBS containing 0.1% sodium azide (NaN<sub>3</sub>) as a preservative. A highly purified grade of BSA has been added as a stabilizing protein to bring the final protein concentration to 4-5 mg/ml after conjugation.

### **STORAGE/STABILITY:**

Store at 4°C. **DO NOT FREEZE.** Avoid 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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**SPECIFICATIONS:**

Clone: D7

Speificity: Mouse Ly-6A/E (Sca-1)

Ig Class: Rat IgG<sub>2a</sub>

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. Re-suspend 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  $\sim 0.25 \mu\text{g}^*$  of **CL8948PE or CL8948PE-3**.
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 tubes are protected from light as most fluorochromes are light sensitive.)
7. Wash 2 times at 4°C.
8. Re-suspend 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 by Flow Cytometry Analysis:

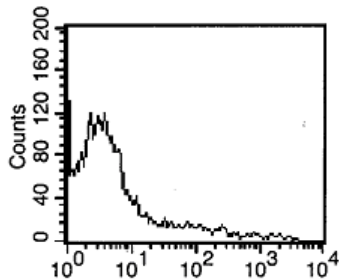
**(Representative Histogram)**

Mouse Strain: C57BL/6

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

Antibody Concentration used:  $\sim 0.25 / 10^6$  cells

Isotypic Control: R-PE Rat IgG2a (CLCR2A04)



Cell Source: mouse bone marrow cells

**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. Ortega, G., P.E. Kerty, E.M. Sherach, and T.R. Malek. 1986. Role of Ly-6 in lymphocyte activation. I. Characterization of a monoclonal antibody to a nonpolymorphic Ly-6 specificity. *J. Immunol.* **137**: 3240-3246.
2. Palfree, R.G.E., F.J. Dumont, and U. Hammerling. 1986. Ly-6A.2 and Ly-6E.1 molecules are antithetical and identical to MALA-1. *Immunogenetics* **23**: 197-207.
3. Codias, R., J. Rutter, T. Fleming, and T. Malek. 1990. *J. Immunol.* **145**:1407.
4. Szilvassy, S. J. and S. Corey. 1993. *Blood.* **81**:2310.
5. Moore, T, M. Bennett and V. Kumar. 1995. *J. Immunol.* **154**:1653.

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