

## Monoclonal Antibody to CD8 - PE

<b>Alternate names:</b>	CD8 alpha chain, CD8A, MAL, T-cell surface glycoprotein CD8 alpha chain, T-lymphocyte differentiation antigen T8/Leu-2
<b>Catalog No.:</b>	CL008R
<b>Quantity:</b>	50 µg
<b>Concentration:</b>	0.1 mg/ml
<b>Background:</b>	The CD8 antigen is a cell surface glycoprotein found on most cytotoxic T lymphocytes that mediates efficient cell to cell interactions within the immune system. The CD8 antigen, acting as a coreceptor, and the T cell receptor on the T lymphocyte recognize antigen displayed by an antigen presenting cell (APC) in the context of class I MHC molecules. The functional coreceptor is either a homodimer composed of two alpha chains, or a heterodimer composed of one alpha and one beta chain. Both alpha and beta chains share significant homology to immunoglobulin variable light chains.
<b>Uniprot ID:</b>	<a href="#">P01731</a>
<b>NCBI:</b>	<a href="#">NP_001074579.1</a>
<b>GeneID:</b>	<a href="#">12525</a>
<b>Host / Isotype:</b>	Rat / IgG2a
<b>Clone:</b>	CT-CD8a
<b>Format:</b>	<b>State:</b> Liquid purified IgG <b>Buffer System:</b> PBS, 0.1% sodium azide (NaN <sub>3</sub> ) 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>	Flow Cytometry Analysis (see Protocols). (Reported to be useful in immunohistochemistry on acetone-fixed frozen sections.) Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
<b>Specificity:</b>	This anti-mouse CD8a antigen monoclonal antibody recognizes the mouse CD8α chain. The α chain of CD8 associates with the CD8β chain to form a CD8α/β heterodimer that is expressed by the majority of thymocytes and by the MHC class I restricted subset of mature T cells <sup>1</sup> . Mouse CD8α can also form a CD8α/α chain homodimer on subsets of CD8 positive cells. For this reason, antibodies specific for CD8a rather than CD8b are recommended for a rigorous delineation of CD8 positive cells. <b>Species:</b> Mouse. Other species not tested.

**For research and in vitro use only. Not for diagnostic or therapeutic work.**

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**Storage:** Store the antibody undiluted at 2-8°C.  
DO NOT FREEZE!  
This product is photosensitive and should be protected from light.  
Shelf life: one year from despatch.

**General References:** 1. Tomonari, k. and Spencer, S. 1990 Epitope-specific binding of CD8 regulates activation of T cells and induction of cytotoxicity. *International Immunology* 2(12): 1189-1194.  
2. Sharon, M., et al. 1999. Interleukin-12 Gene Transfer Results in CD8-Dependant Regression of Mouse CT26 Liver Tumors. *Animals of Surgical Oncology* 6(2): 186-194.

**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  $\sim 1.0 \mu\text{g}$  of this Ab per  $1 \times 10^6$  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).

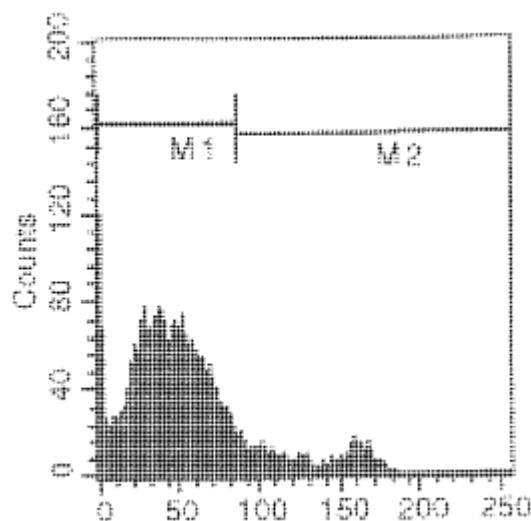
**Results:**

Mouse Strain: BALB/c

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

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

**Pictures:**



Representative Histogram - Isotypic Control: PE Rat IgG2a Percentage of cells stained above control: 13.04%

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