

Product datasheet for **CL009P**

Cd8a Mouse Monoclonal Antibody [Clone ID: 49-31.1]

Product data:

Product Type:	Primary Antibodies
Clone Name:	49-31.1
Applications:	CT, FC
Recommended Dilution:	Flow Cytometry.
Reactivity:	Mouse
Host:	Mouse
Isotype:	IgG3
Clonality:	Monoclonal
Immunogen:	<u>Recipient:</u> 129/ReJ <u>Donor:</u> CBA <u>Fusion Partner:</u> Spleen from immunized recipient fused with Myeloma P3 NSI-Ag 4-1
Specificity:	Anti-mouse Ly-2.1 monoclonal antibody reacts with a sub-population of lymphocytes from mouse strains expressing the Ly 2.1 (CD8a) phenotype, but does not react with lymphocytes from mouse strains expressing the Ly 2.2 phenotype.
Formulation:	PBS containing 0.02% sodium azide (NaN ₃) as preservative State: Purified State: Liquid purified Ig fraction
Concentration:	lot specific
Purification:	Affinity chromatography on Protein G
Conjugation:	Unconjugated
Storage:	Store the antibody undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer. Avoid repeated freezing and thawing.
Stability:	Shelf life: one year from despatch.
Gene Name:	CD8 antigen, alpha chain
Database Link:	<u>Entrez Gene 12525 Mouse P01731</u>
Synonyms:	CD8 alpha chain, CD8A, MAL



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Note: Protocol: **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 2×10^7 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 0.1 - 0.05 μ g of this antibody.
5. Vortex the tubes to ensure thorough mixing of antibody and cells.
6. Incubate the tubes for 30 minutes at 4°C.
7. Wash 2 times at 4°C.
8. Add 100 μ l of secondary antibody (FITC Goat anti-mouse IgG-3 (H+L)) at 1/500 dilution.
9. Incubate the tubes at 4°C for 30-60 minutes.
(It is recommended that the tubes are protected from light since most fluorochromes are light sensitive).
10. Wash 2 times at 4°C in media B.
11. Resuspend the cell pellet in 50 μ l ice cold media B.
12. 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: C3H/He

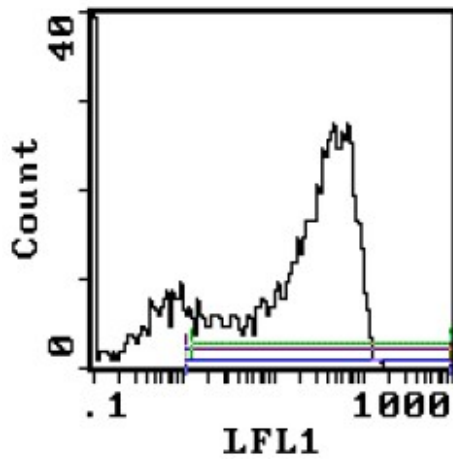
Cell Concentration: 1×10^6 cells per test

Antibody Concentration Used: 0.1 μ g/ 10^6 cells

Isotypic Control: Purified Mouse IgG-3

Percentage of cells stained above control:
Thymus 78.9%

Product images:



Strain Distribution by Flow Cytometry Analysis:

Procedure: As above

Antibody Concentration: 0.1 $\mu\text{g}/10^6$ cells

Strains tested:

<u>Strain</u>	<u>Phenotype</u>	<u>+/-</u>
C57BL/6	Ly-2.2	-
CBA/J	Ly-2.1	+
Balb/c	Ly-2.2	-
C3H/He	Ly-2.1	+