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Product datasheet for CL008P

Cd8a Rat Monoclonal Antibody [Clone ID: CT-CD8a]

Product data:

Product Type:	Primary Antibodies
Clone Name:	CT-CD8a
Applications:	FC, IHC
Recommended Dilution:	Flow Cytometry Analysis (See Protocol). (Reported to be useful in immunohistochemistry on acetone fixed frozen sections.)
Reactivity:	Mouse
Host:	Rat
Isotype:	IgG2a
Clonality:	Monoclonal
Specificity:	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 cells1. 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.
Formulation:	PBS and 0.02% Sodium Azide as preservative. State: Purified State: Liquid purified IgG fraction.
Concentration:	lot specific
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:	Entrez Gene 12525 Mouse P01731

Background: 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.

Synonyms: CD8 alpha chain, CD8A, MAL

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 2x10e7 cells/ml in media A. Add 50 µl of this suspension to each tube (each tube will then contain 1x10e6 cells, representing 1 test).
4. To each tube, add ~1.0 µg* of this Ab.
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 µl of secondary antibody (FITC Goat anti-rat IgG (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 µl ice cold media B.
12. Transfer to suitable tubes for flow cytometric analysis containing 15 µl 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 µl of 2M sodium azide in 100 mls).
B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 µl of 2M sodium azide in 100 mls).