

Product datasheet for CL001BX

OriGene Technologies, Inc.

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Cd3e Hamster Monoclonal Antibody [Clone ID: 145-2C11]

Product data:

Product Type: Primary Antibodies

Clone Name: 145-2C11

Applications: FC

Recommended Dilution: Flow Cytometry (See Protocols).

This clone has also been reported to work in Immunoprecipitation ^{1,2} and Western

Blotting 8.

Reactivity: Mouse Hamster

Isotype: IgG

Clonality: Monoclonal

Immunogen: H-2Kb sp from Armenian Hamster Spleen.

Fusion Partner: Murine myeloma cell line SP2/0

Specificity: This anti-Mouse CD3e monoclonal antibody is specific for a 25 kDa protein component (e-T3)

of the antigen specific T cell receptor on all mouse strains tested. The e-T3 protein has been shown to be non-covalently associated on the cell surface ab heterodimer of the CD3 associated complex. This monoclonal antibody reacts with all mature T cells and can both activate and inhibit T cell function (1). This fact identifies e-T3 as a cell surface protein involved in the transduction of activation signals. All peripheral T cells express this

determinant however B cells and bone marrow cells have proven to be negative. Although the expression of this particular epitope on peripheral T cells is uniformly high, staining of thymocytes reveals distinct subpopulations of cells differing in the level of expression of this

marker.

This antibody will prove useful in studying the role of various components of the TCR complex in T cell activation and development, and will allow for the development of an animal model in which to investigate the immunoregulatory effects of *in vivo* administration of anti-CD3 antibodies, an area of obvious clinical importance. Anti-CD3e is ideal for flow cytometry applications, particularly as a specific marker for tracking mouse T cells. In addition, this monoclonal antibody, clone 145-2C11, was specifically designed to trigger T cell

activation.





Cd3e Hamster Monoclonal Antibody [Clone ID: 145-2C11] - CL001BX

Formulation: PBS, 0.2 μm sterile filtered

Label: Biotin

State: Liquid purified Ascites

Concentration: lot specific

Purification: Protein G Affinity Chromatography

Conjugation: Biotin

Storage: Store 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: CD3 antigen, epsilon polypeptide

Database Link: Entrez Gene 12501 Mouse

P22646

Background: T cell activation through the antigen receptor (TCR) involves the cytoplasmic tails of the CD3

subunits: CD3 gamma, CD3 delta, CD3 epsilon and CD3 zeta. These CD3 subunits are

structurally related members of the immunoglobulins super family encoded by closely linked genes on human chromosome 11. The CD3 components have long cytoplasmic tails that associate with cytoplasmic signal transduction molecules. This association is mediated at least in part by a double tyrosine based motif present in a single copy in the CD3 subunits. CD3 may play a role in TCR induced growth arrest, cell survival and proliferation. The CD3 antigen is present on 68-82% of normal peripheral blood lymphocytes, 65-85% of thymocytes and

Purkinje cells in the cerebellum. It is never expressed on B or NK cells. Decreased percentages of T lymphocytes may be observed in some autoimmune diseases.

Synonyms: T3/Leu-4



Note:

Donor: BALB/c

Cell Concentration: 1x106 cells

Antibody Concentration: 1 µg/106 cells <u>Isotypic Control</u>: Biotin Hamster IgG

Protocol: Flow Cytometry Analysis:

- 1. Prepare 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 cells to $1x10^6$ cells in approximately 50 μ l Media A in a microcentrifuge tube (i.e. 50 μ l of cells resuspended to $2x10^7$ cells/ml). (THE CONTENTS OF 1 TUBE REPRESENTS 1 TEST).
- 4. To each tube add 1 μg of this antibody per 10 cells*.
- 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 at a 1/500 dilution.
- 9. Incubate 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 μ l of propidium iodide at 0.5 mg/ml in phosphate buffered saline. (This stains dead cells by intercalating DNA).

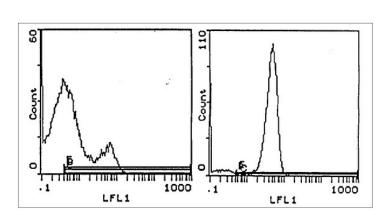
Media:

- A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide ($100 \mu l$ of 2 M sodium azide in 100 mls).
- B. Phosphate buffered saline (pH 7.2) + 0.5% bovine serum albumin + sodium azide (100 μ l of 2 M sodium azide in 100 mls).



Product images:





Cell Source: A/Thymocytes, B/Splenic T Cells Percentage of Cells Stained Above Control: A/50.2, B/87.6