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## **Biotin Anti-Mouse Dendritic Cells (Cell Surface) Monoclonal Antibody**

**CL89145B**

**Lot: 14541**

### **DESCRIPTION:**

CL89145F (NLDC-145) identifies Ia positive interdigitating cells, veiled cells, and Langerhans cells of the skin and their *in vitro* counterparts. The antigen, also known as DEC-205, is expressed at high levels by dendritic cells and thymic epithelial cells. The dendritic cells that express high levels of the antigen are those in the skin, in the T cell regions of peripheral lymphoid organs and dendritic cells that are grown from proliferating bone marrow precursors in the presence of high dose GM-CSF. DEC-205 can also be expressed on B cells, although at much lower levels. It is absent in freshly isolated macrophages from the peritoneal cavity although a sub-population becomes weakly positive in mice previously stimulated with thioglycollate. The antigen detected by CL89145 is an integral membrane glycoprotein with an apparent mass of 205 kDa. DEC-205 is apparently a receptor involved in antigen-processing by dendritic cells. The antigen recognized by CL89145 is localized in the cytoplasm (after fixation) and on the cell surface. It is more widely distributed than the CL89148 (MIDC-8) antigen which is a very specific cytoplasmic component. This clone can be used in flow cytometry, immunohistochemistry, immunocytochemistry, immunoblotting and for functional studies.

### **PRESENTATION:**

100 µg biotin conjugated Ig buffered in PBS , 0.02% NaN<sub>3</sub> and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml.

### **STORAGE/STABILITY:**

Store at 4°C. For long term storage, aliquot and freeze unused portions at -20°C in volumes appropriate for single usage. Avoid freeze/thaw cycles.

For more information or to place an order please contact...

**CEDARLANE®**  
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**SPECIFICITY:** Reacts with interdigitating cells, dendritic cells.

**CLONE:** NLDC-145

**ISOTYPE:** Rat IgG<sub>2a</sub>

**FORMAT:** Biotin conjugated Ig buffered in PBS, 0.02% NaN<sub>3</sub> and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml. (Affinity Purified IgG from culture supernatant)

**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<sup>®</sup>-Mouse cell separation medium (CL5030).
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 1.0-0.5  $\mu$ g\* of **CL89145B**.
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  $\mu$ l of secondary antibody **CLCSA1004** (PE-Streptavidin) 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  $\mu$ l ice cold media B.
12. 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).

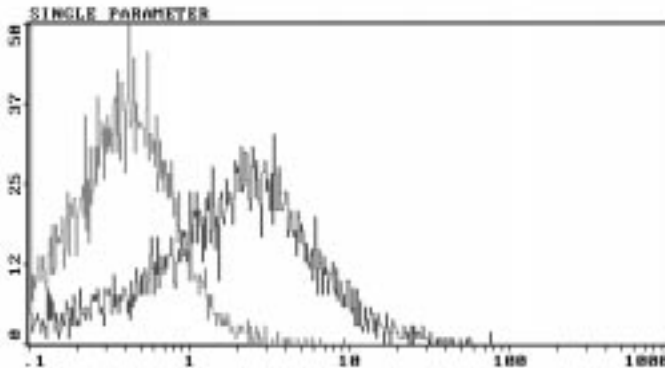
**RESULTS:**Tissue Distribution by Flow Cytometry Analysis:

Mouse Strain: BALB/c

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

Antibody Concentration Used:  $0.5 \mu\text{g}/10^6$  cells

Isotypic Control: Biotin Rat IgG<sub>2a</sub>



LFL2

Cell Source: Dendritic Cells

Percentage of cells stained above control: 39.5 %

**N.B. Appropriate control samples should always be included in any labeling studies.**

**\* For optimal results in various applications, it is recommended that each investigator determine dilutions appropriate for individual use.**

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