

## Product datasheet for **CL121BX**

### MHC Class II RT1D Mouse Monoclonal Antibody [Clone ID: OX-17]

#### Product data:

|                       |   |
|-----------------------|---|
| Product Type:         | Primary Antibodies  |
| Clone Name:           | OX-17   |
| Applications:         | FC, IHC   |
| Recommended Dilution: | Flow Cytometry.   |
| Reactivity:           | Rat   |
| Host:                 | Mouse   |
| Isotype:              | IgG1  |
| Clonality:            | Monoclonal  |
| Immunogen:            | Rat spleen membrane glycoproteins depleted of Ia-A antigens.<br>Immunocyte Donor: BALB/c spleen<br>Fusion Partner: X63 Ag8.653  |
| Specificity:          | This monoclonal antibody recognizes a monomorphic determinant on the a chain of the rat Ia antigen and appears to be the rat homologue of mouse Ia-E. It recognizes the rat Ia product present on B, but not T cells from lymph node or thoracic duct lymph. It does not bind to thymocytes or erythrocytes. The antibody does not cross-react with rat Ia-A or mouse Ia-E antigen, but rabbit antibody raised against the antibody affinity column-purified MRC OX-17 antigen cross-reacted on tissues of mice expressing Ia-E mouse antigen but not on those mouse strains that were Ia-E antigen negative. |
| Formulation:          | 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<br>Label: Biotin<br>State: Liquid purified Ig  |
| Concentration:        | lot specific  |
| Purification:         | Protein G Chromatography  |
| Conjugation:          | Biotin  |
| Storage:              | Store the antibody undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer.<br>Avoid repeated freezing and thawing.   |
| Stability:            | Shelf life: one year from despatch.   |



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**Synonyms:** HLA Class II

**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®-Rat 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 0.5-0.1  $\mu$ g\* of this Ab per  $10^6$  cells.
5. Vortex the tubes to ensure thorough mixing of antibody and cells.
6. Incubate the tubes for 30 minutes at 4°C.
7. Wash 2 times at 4°C.
8. Add 100  $\mu$ l of secondary antibody (Streptavidin-FITC) at a 1:500 dilution.
9. Incubate tubes at 4°C for 30 - 60 minutes (It is recommended that tubes are protected from light since most fluorochromes are light sensitive).
10. Wash 2 times at 4°C.
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:**

Rat Strain: Fischer

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

Antibody Concentration Used: 0.1  $\mu$ g/ $10^6$  cells

Isotypic Control: Biotin Mouse IgG1

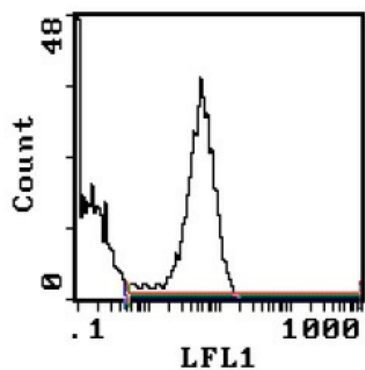
**Cell Source Percentage of cells stained above control:**

Thymus 6.1%

Spleen 48.8%

Lymph Node 27.5%

## Product images:



Cell Source: Spleen

Percentage of cells stained above control: 48.8%