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Product datasheet for CL121P

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

Product data:

Product Type:	Primary Antibodies
Clone Name:	OX-17
Applications:	FC, IHC, IP
Recommended Dilution:	Flow Cytometry: 1/250 - 1/500 (see Protocols). Immunhistochemistry. Immunoprecipitation.
Reactivity:	Rat
Host:	Mouse
Isotype:	IgG1
Clonality:	Monoclonal
Immunogen:	Rat spleen membrane glycoproteins depleted of Ia-A antigens Immunocyte Donor: BALB/c spleen Fusion Partner: X63 Ag8.653
Specificity:	This monoclonal antibody recognizes a monomorphic determinant on the α 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 with 0.02% sodium azide State: Purified State: Liquid purified IgG
Concentration:	lot specific
Purification:	Protein G affinity chromatography
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.

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Synonyms: HLA Class II

Note: Protocol: **FLOW CYTOMETRY ANALYSIS:**

METHOD:
1. Prepare 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 cells to 1x10e6 cells in approximately 50 µl media A in a microcentrifuge tube. (i.e. 50 µl of cells resuspended to 2x10e7 cells/ml). The contents of 1 tube represent 1 test.
4. To each tube add 50 µl of a 1:250 - 1:500 dilution 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-mouse IgG (H+L)) at 1:700 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 µl 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 µl of 2 M sodium azide in 100 mls).
B. Phosphate buffered saline (pH 7.2) + 0.5% bovine serum albumin + sodium azide (100 µl of 2 M sodium azide in 100 mls).

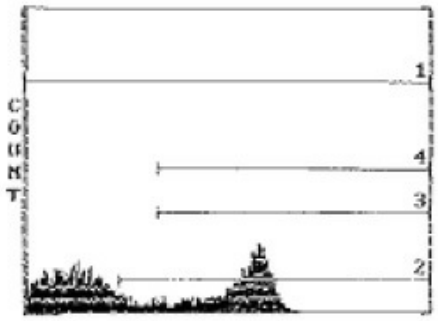
RESULTS:
Rat Strain: Lewis Rat
Cell Concentration: 1x10e6 cells per test
Antibody Concentration: 1:400
Isotypic Control: Mouse IgG1, κ

CELL SOURCE PERCENT STAINING
Thymus 12%
Spleen 34%
Lymph Node 25%

STRAIN DISTRIBUTION:
Antibody Concentration: 1:100
Strains Tested: Wistar, Buffalo, Brown Norway, Fischer 344
Positive: Wistar, Buffalo, Brown Norway, ACI, Fischer 344

Negative: none

Product images:



LFL1
Cell Source: Splenocytes
Percentage of Cells Stained Above Control: 34%