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Product datasheet for SM2201B

HLA Class II DR Rat Monoclonal Antibody [Clone ID: YD1/63.4.10]

Product data:

Product Type:	Primary Antibodies
Clone Name:	YD1/63.4.10
Applications:	FC
Recommended Dilution:	**Flow Cytometry.** **Immunohistochemistry on Cryostat and Paraffin-Embedded Sections.**
Reactivity:	Human
Host:	Rat
Isotype:	IgG2a
Clonality:	Monoclonal
Immunogen:	DAUDI cells. **Donor:** immunized DA rat spleen cells. **Fusion Partner:** Y3 Ag1.2.3 rat myeloma.
Specificity:	Anti-Human HLA-DR monoclonal antibody recognizes the HLA-DR (MHC class II) antigen.
Formulation:	PBS containing 0.02% Sodium Azide as preservative and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml. Label: Biotin State: Liquid purified Ig fraction
Concentration:	lot specific
Purification:	Affinity Chromatography on Protein G
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.
Synonyms:	HLA-DR, HLA class II histocompatibility antigen DR, MHC class II antigen DR

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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®-H 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 50µl of a 0.5-0.2 µg dilution of this antibody per 10e6 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 (Streptavidin-FITC) at a 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).

Results:

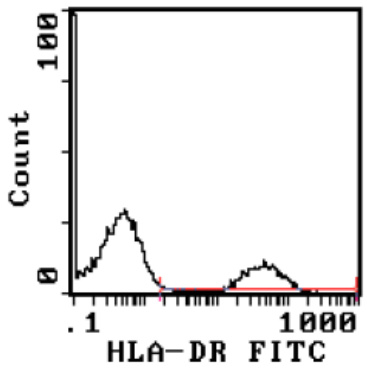
Tissue Distribution by Flow Cytometry Analysis:

Cell Concentration: 1x10e6 cells per test

Antibody Concentration Used: 0.5 µg/10e6 cells

Isotypic Control: Biotin Rat IgG2a

Product images:



Cell Source: Peripheral Blood Lymphocytes
Percentage of cells stained above control: 26.7%