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- Diagnostik & molekulare Diagnostik
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TECHNICALLY Speaking

Place your order with CEDARLANE® or your local distributor.

Please contact CEDARLANE® for lot specific information.

Purified Anti-Rat RT1.B Monoclonal Antibody

CL010AP
CL010AP-2
Lot: 1021

DESCRIPTION:

Cedarlane's anti-rat RT1.B monoclonal antibody recognizes Ia from all rat strains tested. It labels a monomorphic determinant of the rat Ia antigen present on the majority of peripheral B lymphocytes, dendritic cells, some macrophages, and certain epithelial cells (2,3,4) but not peripheral T lymphocytes. (4) This antibody also cross reacts with murine Ia antigens (particularly with k and s haplotypes). Analysis of recombinant strains shows that the determinant maps to the I-A region of the mouse MHC (1,2) and correlates with mouse Ia specificity 17 or 18. Thus, the determinant recognized by CL010AP is polymorphic in the mouse. This antibody may be useful for recognizing the rat Ia antigen and can be coupled to Sepharose 4B for purification of rat Ia antigen by antibody affinity chromatography (1).

This clone is reported to work with frozen and paraffin sections (5).

PRESENTATION:

250 µg (CL010AP) or 500 µg (CL010AP-2) purified Ig buffered in PBS + 0.02% NaN_3.

STORAGE AND RECONSTITUTION:

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

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

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SPECIFICATIONS:

CLONE: MRC OX-6

HYBRIDOMA PRODUCTION:

IMMUNIZATION:

IMMUNOGEN: Rat thymocyte membrane glycoprotein.
IMMUNOCYTE DONOR: BALB/c spleen
FUSION PARTNER: P3-NS1-1-Ag4 (NS1/1)
SPECIFICITY: Rat RT1.B (Rat Ia-non-polymorphic)
Ig CLASS: Mouse IgG_1
PRESENTATION: Purified Ig in PBS with the addition of 0.02% NaN_3.
(Ascites purified via Protein G Chromatography)

ANTIBODY CONCENTRATION: 1.0 mg/ml

FLOW CYTOMETRY ANALYSIS:

1. Prepare cell suspension in media A. For cell preparations, deplete the red blood cell population with Lympholyte ® Rat cell separation medium (CL5040).
2. Wash 2 times.
3. Resuspend cells to 1 x 10^6 cells in approximately 50 µl media A in a microcentrifuge tube. (ie. 50 µl of cells resuspended to 2 x 10^7 cells / ml).
   the contents of 1 tube represent 1 test.
4. To each tube add 50 µl of a 1:250-1:1000 dilution of **CL010AP or CL010AP-2**.
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 **CLCC30201** (Goat anti-mouse IgG (H+L)-FITC conjugate) 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

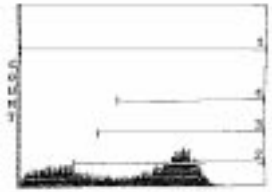
A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide (100 µl of 2M sodium azide in 100 mls).
B. Phosphate buffered saline (pH 7.2) + 0.5% bovine serum albumin + sodium azide (100 µl of 2M sodium azide in 100 mls).

FLOW CYTOMETRIC ANALYSIS

Rat Strain: Wistar Rat
Cell Concentration: 1 X 10^6 cells per test
Antibody Concentration: 1:400
Isotypic Control: Mouse IgG_1, k

***RECOMMENDED DILUTION FOR USE:** 1:250 -1:1000

CELL SOURCE	PERCENT STAINING
Thymus	8.2%
Spleen	48.1%
Lymph Node	21.8%



LFL1
Cell Source: Splenocytes
Percentage Of Cells Stained Above Control: 48.1%

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

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

STRAIN DISTRIBUTION:
PROCEDURE: as above
ANTIBODY CONCENTRATION: 1:500
STRAINS TESTED: Wistar, Buffalo, Brown Norway, Fischer , Lewis

POSITIVE: Wistar, Buffalo, Fischer , Lewis, Brown Norway
NEGATIVE: none

REFERENCES:

1. McMaster, W.R. and A.F. Williams. (1979) Identification of Ia glycoproteins in rat thymus and purification from rat spleen. Eur. J. Immunol. 9, 426-433.
2. Barclay, A.N. (1981) The localization of populations of lymphocytes defined by monoclonal antibodies in rat lymphoid tissues. Immunology. 42, 593-600.
3. Barclay, A.N. and G. Mayrhofer. (1981) Bone Marrow origin of Ia-positive cells in the medulla of rat thymus. J. Exp. Med. 153, 1666-1671.
4. Barclay, A.N. (1981) Different reticular elements in rat lymphoid tissue identified by localization of Ia, Thy-1 and MRC OX-2 antigens. Immunology. 44, 727-736.
5. Whiteland, J.L et al (1995). Immunohistochemical detection of T cell subsets and other leukocytes in paraffin embedded rat and mouse tissues with monoclonal antibodies .J. Histochem. Cytochem. 43: 313-320.

FOR LABORATORY RESEARCH ONLY

(Not to be administered to humans or animals nor used for any drug purpose).