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TECHNICALLY Speaking

Place your order with CEDARLANE® or your local distributor.

Please contact CEDARLANE® for lot specific information.

Biotin Anti-Human HLA-DR Monoclonal Antibody

CLHLA-03B
CLHLA-03B-3
LOT: 0341

DESCRIPTION:

Cedarlane's Biotin anti-human HLA-DR monoclonal antibody recognizes the HLA-DR (MHC class II) antigen.

Applications include: flow cytometry, cryostat sections, Paraffin-embedded sections

PRESENTATION:

100µg (CLHLA-03B) or 300µg (CLHLA-03B-3) purified Ig buffered in PBS with 0.02% NaN_3 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 portion at -20°C in volumes appropriate for single usage. Avoid repeated freeze/thaw cycles as this may denature the antibody.

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

CEDARLANE® LABORATORIES LIMITED

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in North America

phone: (905) 878-8891 • fax: (905) 878-7800

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or visit our website for a list of our international distributors including contact information

website: www.cedarlanelabs.com • e-mail: info@cedarlanelabs.com

SPECIFICATIONS:

Clone: YD1/63.4.10

Hybridoma Production:

Immunization: Immunogen: DAUDI cells
Donor: immunized DA rat spleen cells

Fusion Partner: Y3 Ag1.2.3 rat myeloma

Specificity: HLA-DR antigen

Ig Class: Rat IgG_{2a}

Format: Biotin conjugated Ig buffered in PBS, 0.02% NaN_3 and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml. (Purified from ascites via Protein G Chromatography)

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®-H (CL5010) cell separation medium.
2. Wash 2 times.
3. Resuspend the cells to a concentration of $2x10^7$ cells/ml in media A. Add 50 µl of this suspension to each tube (each tube will then contain $1x10^6$ cells, representing 1 test).
4. To each tube, add 50µl of a 0.5-0.2µg dilution of **CLHLA-03B** or **CLHLA-03B-3** 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 µl of secondary antibody **CLCSA1001** (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.

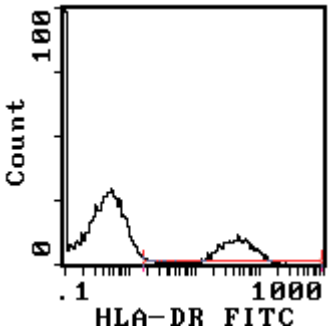
Media:

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 : $1x10^6$ cells per test
Antibody Concentration Used: 0.5 µg/10^6 cells
Isotypic Control: Biotin Rat IgG_{2a} (CLCR2a15)



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

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.**

REFERENCES:

1. Janossy, G., Tidman, N., Crawford, D., Papageargiou, E. S., Prentice, H. G., Francis, G., Bradstock, K. F., McConnell, I., Secher, D., and Milstein, C. (1980). Protides of Biol. Fluids 28: 523-528

FOR RESEARCH USE ONLY

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