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Monoclonal Antibody to CD45 / LCA (CD45RB) - Purified

Alternate names: L-CA, Leukocyte common antigen, PTPRC, Receptor-type tyrosine-protein phosphatase C, T200

Catalog No.: CL029P

Quantity: 0.25 mg

Concentration: 1.0 mg/ml

Background: CD45 is a family of single chain transmembraneous glycoproteins consisting of at least four isoforms (220, 205, 190, 180 kDa) which share a common large intracellular domain. Their extracellular domains are heavily glycosylated. The different isoforms are produced by alternative messenger RNA splicing of three exons of a single gene on chromosome 1. CD45 is expressed on cells of the human hematopoietic lineage (including hematopoietic stem cells) with the exception of mature red cells. It is not detected on differentiated cells of other tissues. It is likely that CD45 plays an important role in signal transduction, inhibition or upregulation of various immunological functions. Antibodies recognising a common epitope on all of the isoforms are termed CD45 whilst those recognising only individual isoforms are termed CD45RA or CD45RO etc.

Uniprot ID: P06800

NCBI: 10090

Host / Isotype: Rat / IgG2a

Clone: 16A

Immunogen: TH2 cell clones, final boost with TH2 clone D10

Format:
State: Liquid purified Ig fraction
Purification: Protein G chromatography
Buffer System: PBS containing 0.02% sodium azide

Applications: Immunohistochemistry on acetone-fixed frozen sections and paraffin embedded sections.
Immunoprecipitation.
Flow cytometry (for details see "Specificity" and "Protocols" below).
Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

For research and in vitro use only. Not for diagnostic or therapeutic work.
Material Safety Datasheets are available at www.acris-antibodies.com or on request.

Specificity:

Anti-CD45RB monoclonal antibody reacts with the CD45 isoform containing the exon B dependent epitope.
CD45RB is highly expressed on peripheral B cells, cytotoxic T cells, a subset of T helper cells and most thymoctyes.
Tissue distribution by flow cytometry analysis:
Mouse strain: BALB/c
Cell concentration : 1x10e6 cells per test
Antibody dilution used: 50 µl at 1/5000/10e6 cells
Isotypic control: Rat IgG2a
Cell source / Percentage of cells stained above control:
Thymus / 81.1%
Spleen / 63.0%
Lymph Node / 74.1%
Species: Mouse.
Other species not tested.

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.
Shelf life: one year from despatch.

General References:

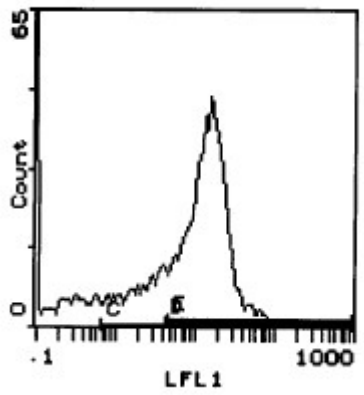
1. Bottomly, K., M. Lugman, L. Greenbaum, S. Carding, I. West, T. Pasqualini, and D.B. Murphy. A monoclonal antibody to murine CD45R distinguishes CD4 T cell populations that produce different cytokines. Eur. J. Immunol.1989. 19:617-623.
2. Hathcock, K.S., H. Hirano, R.J. Hodes. CD45 expression by murine B cells and T cells: Alteration of CD45 isoforms in subpopulations of activated B cells. Immunol. Res. 1993. 12:21-36.

Protocols:

FLOW CYTOMETRY ANALYSIS:
Method:
1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population
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 50 µl of a 1/2500-1/5000 dilution of CL029P.
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-rat IgG (H+L)) 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).

Pictures:



Cell source: Spleen.
Percentage of cells stained above control: 63.0%