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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

TECHNICALLY *Speaking*

Place your order with CEDARLANE® or your local distributor.

Please contact CEDARLANE® for lot specific information.

Anti-Rabbit Pan Leukocytes (CD58) Monoclonal Antibody-Ascites

**CL8804A
LOT: 8411**

DESCRIPTION:

Cedarlane's anti-rabbit pan leukocytes (CD58) monoclonal antibody recognizes an antigen that appears to be the rabbit equivalent of CD58/LFA-3 present on all leukocytes as well as erythrocytes (1). The level of expression of this antigen differs markedly from population to population with the highest levels on monocytes, peritoneal macrophage, moderate levels on neutrophils and low levels on T and B cells and platelets.

Applications include: flow cytometry and immunoprecipitation.

PRESENTATION: 0.5 ml, lyophilized ascites

STORAGE/STABILITY:

Lyophilized form stable at 4°C or -20°C. Reconstitute with 0.5 ml of cold distilled water. After reconstitution, aliquot and freeze unused portions in volumes appropriate for single usage. Avoid repeated freeze/thaw cycles.

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

CEDARLANE®
LABORATORIES LIMITED



toll free: 1-800-268-5058
in North America

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

5516 - 8th Line, R.R.#2, Hornby, Ontario, CANADA L0P 1E0

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: VC21

Hybridoma Production:

Immunization: Immunogen: Veiled rabbit cells
Donor: BALB/c spleen

Fusion Partner: Myeloma cell line SP2/0

Specificity: Rabbit Pan Leukocyte (CD58)

Ig Class: Mouse IgG₁

Format: Ascitic fluid, filtered to 0.8 μ (non-sterile) and lyophilized.

FLOW CYTOMETRY ANALYSIS:**Method:**

1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population with Lympholyte®-Rabbit cell separation medium (CL5050).
2. Wash 2 times.
3. Resuspend the cells to a concentration of 2×10^7 cells/ml in media A. Add 50 μl of this suspension to each tube (each tube will then contain 1×10^6 cells, representing 1 test).
4. To each tube, add 50 μl of a 1:500-1:5000 dilution * of **CL8804A**.
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** (FITC Goat anti-mouse IgG (H+L)) at 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 μ l of 2M sodium azide in 100 mls).
- B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 μ l of 2M sodium azide in 100 mls).

Results:Tissue Distribution by Flow Cytometry Analysis:

Cell Concentration : 1×10^6 cells per tests

Antibody Concentration Used: 1:4000 in 50 μ l / 10^6 cells

Isotypic Control: Mouse IgG₁

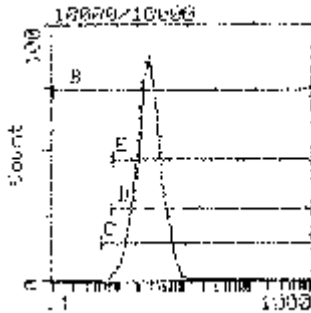
Cell SourcePercentage of cells stained above control:

Thymus

99.5%

Spleen

94.3%



LFL1

Cell Source: Thymus

Percentage of cells stained above control: 99.5%

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 dilution's appropriate for individual use.**

REFERENCES:

- 1) Wilkinson, J.M., Galea-Lauri, J., Sellars, R.A., Boniface, C. 1992. Identification and tissue distribution of rabbit leucocyte antigens recognized by monoclonal antibodies. *Immunology* 76:625-630.

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