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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.

Purified Anti-Rat CD43 Monoclonal Antibody NO AZIDE

CL002NA

DESCRIPTION:

Cedarlane's anti-rat CD43 monoclonal antibody recognizes a monomorphic determinant expressed on rat thymocytes, polymorphonuclear cells, plasma cells and stem cells, but not B lymphocytes or pre-B cells (1,3). The antigen is a heavily glycosylated glycoprotein of apparent molecular weight 95,000 and has a high content of O-linked carbohydrate structures (3). This major glycoprotein of thymocytes and T lymphocytes is referred to by several names including leukocyte sialoglycoprotein and leukosialin. The carbohydrate structures of leukosialin account for approximately 60% of its weight (2). On thymocytes, this glycoprotein is the main target for binding of peanut lectin (4). This antibody is useful for labelling T but not B lymphocytes and in studies on stem cells since pre-B cells are not labelled while the multipotential stem cell is. It may also be used in analysis of NK cells (5) and in molecular studies in the sialoglycoprotein which it recognizes. This clone has been reported to work in immunohistochemistry (frozen sections).

PRESENTATION:

1.0 mg (CL002NA) purified Ig buffered in PBS, **No Preservative.**

STORAGE/STABILITY:

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

Continued Overleaf...

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
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SPECIFICATIONS:

Clone: W3/13 HLK

Hybridoma Production:

Immunization: Immunogen: Rat thymocyte membrane
Immunocyte Donor: BALB/C spleen

Fusion Partner: NS1/1

Specificity: Rat CD43

Isotype: Mouse IgG₁

Presentation: Purified Ig buffered in PBS, No Preservative. (Purified from ascitic fluid 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 1×10^6 cells in approximately 50 μ l Media A in a microcentrifuge tube. (i.e. 50 μ l of cells resuspended to 2×10^7 cells/ml.)
(The Contents Of 1 Tube Represent 1 Test.)
4. To each tube add 1.0 – 0.5 μ g* of **CL002NA** 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 μ l of secondary antibody **CLCC30201** (FITC Goat anti-mouse IgG (H+L) at 1:500 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 μ l ice cold media B.
12. Transfer to suitable tubes for flow cytometric analysis containing 15 μ l 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 μ l of 2 M sodium azide in 100 mls.)
- B. Phosphate buffered saline (pH 7.2) + 0.5 % bovine serum albumin + sodium azide (100 μ l of 2 M sodium azide in 100 mls.)

Rat Strain: Fisher

Cell Concentration: 1×10^6 cells per test

Antibody Concentration: 0.5 μ g / 10^6 cells

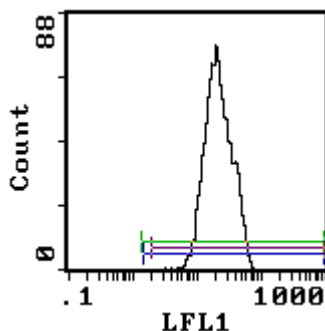
Isotypic Control: Purified Mouse IgG₁ (CLCMG100)

CELL SOURCE

Thymus
Spleen
Lymph Node

PERCENT STAINING

100%
33.3%
58.9%



Cell Source: Thymus
Percentage of Cells Stained Above Control: 100%

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: 1:200

Strains Tested: Lewis, Wistar, ACI, Brown Norway, Fischer 344, Buffalo

Positive: Lewis, Wistar, ACI, BN, Fischer 344, Buffalo

Negative: none

REFERENCES:

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2. Dyer, M.J.S. and S.V. Hunt. (1981) J.Exp.Med. 154, 1164-1177. Characterization by surface W3/13 antigen and radiosensitivity.
3. Brown, W.R.A., Barclay, A.N., Sunderland, C.A. and A.F. Williams. (1981) Nature. 289, 456-460. Identification of glycoprotein-like molecule at the cell surface of rat thymocytes.
4. Brown, W.R.A. and A.F. Williams. (1982) Immunology. 46, 713-726. Lymphocyte cell surface glycoproteins which bind to soybean and peanut lectins.
5. Cantrell, D.A. Robins, R.A. Brooks, C.G. and R.W. Baldwin. (1982) Immunology. 45, 97-103.
6. Killeen, N., Barclay, A.N., Willis, A.C. and A.F. Williams. (1987) The EMBO J., vol.6, #13, 4029-4034. The sequence of rat leukosialin (W3/13 antigen) reveals a molecule with O-linked glycosylation of one third of its extracellular amino acids.

FOR RESEARCH USE ONLY

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