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Product datasheet for CL023F

Cd44 Rat Monoclonal Antibody [Clone ID: KM81]

Product data:

Product Type:	Primary Antibodies
Clone Name:	KM81
Applications:	FC
Recommended Dilution:	Flow cytometry. Immunohistochemistry.
Reactivity:	Mouse
Host:	Rat
Isotype:	IgG2a
Clonality:	Monoclonal
Immunogen:	Bone Marrow Derived Stromal cells (clone BMS2). Donor: Lou/MN Rat. Fusion Partner: SP2/0.
Specificity:	This monoclonal antibody recognizes a 95 kDa glycoprotein found on most hematopoietic cells (1). It is thought to be important in the regulation of migratory properties of lymphocytes during development and the regulation of the interaction with bone marrow stromal cells during hematopoiesis (2,3). CD44 functions as a receptor for hyaluronate, although some cells expressing CD44 do not bind hyaluronate (3,4). This antibody has been shown to inhibit the growth of lymphoid and myeloid cells on long term bone marrow cultures (3). It also blocks the adhesive interactions of B cell hybridomas to a cloned stromal line or to hyaluronate coated dishes (4).
Formulation:	PBS, 0.02% NaN3 and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml. Label: FITC State: Liquid
Concentration:	lot specific
Purification:	Protein G Chromatography
Conjugation:	FITC
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.

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Stability:	Shelf life: one year from despatch.
Gene Name:	CD44 antigen
Database Link:	Entrez Gene 12505 Mouse P15379
Background:	CD44 is a type 1 transmembrane glycoprotein also known as Phagocytic Glycoprotein 1 (pgp 1) and HCAM. CD44 is the receptor for hyaluronate and exists as a large number of different isoforms due to alternative RNA splicing. The major isoform expressed on lymphocytes, myeloid cells, and erythrocytes is a glycosylated type 1 transmembrane protein. Other isoforms contain glycosaminoglycans and are expressed on hematopoietic and non hematopoietic cells. CD44 is involved in adhesion of leukocytes to endothelial cells, stromal cells, and the extracellular matrix.
Synonyms:	LHR, MDU2, MDU3, MIC4, CDw44, Epican, ECMR-III, HUTCH-I, Heparan sulfate proteoglycan, Hermes antigen, Hyaluronate receptor, PGP-1

Note:

Protocol: **FLOW CYTOMETRY ANALYSIS:**

Method:
1. Prepare a cell suspension in media A. For preparations, deplete the red blood cell population with Lympholyte®-M cell separation medium.
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 1 x 10e6 cells, representing 1 test).
4. To each tube, add 1.0 - 0.5 µg* of this Ab per 10e6 cells.
5. Vortex the tubes to ensure thorough mixing of antibody and cells.
6. Incubate the tubes for 30 minutes at 4°C. (It is recommended that the tubes are protected from light, since most fluorochromes are light sensitive.)
7. Wash 2 times at 4°C.
8. Resuspend the cell pellet in 50 µl ice cold media B.
9. 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.

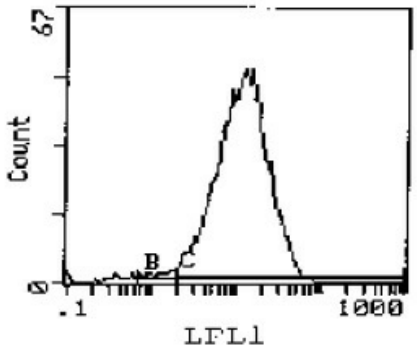
Media:
A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide (100 µl of 2M sodium azide in 100 mls).
B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 µl of 2M sodium azide in 100 mls).

Results - Tissue Distribution:
Mouse Strain: BALB/c
Cell Concentration: 1x10e6 cells per test
Antibody Concentration Used: 0.5 µg/10e6 cells
Isotypic Control: FITC Rat IgG2a

Results - Strain Distribution:
Cell Concentration: 1x10e6 cells per test
Antibody Concentration Used: 0.5 µg /10e6 cells
Strains Tested: BALB/c, CBA/J, C3H/He, C57BL/6, SWR
Positive: BALB/c, CBA/J, C3H/He, C57BL/6, SWR
Negative: none

IMMUNOHISTOCHEMISTRY:
This Anti-mouse CD44 is suitable for staining frozen and acetone-fixed sections with superior staining found on acetone-fixed tissue. The recommended dilution is 1/50 with a minimal incubation time of 1 hour at room temperature.

Product images:



Cell Source: Spleen
Percentage of cells stained above control: 96.2%

Cell Source	Percentage of cells stained above control:
Thymus	57.6%
Spleen	96.2%
Lymph Node	77.4%
Bone Marrow	90.5%