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Product datasheet for CL046F

Ly6g Rat Monoclonal Antibody [Clone ID: RB6-8C5]

Product data:

Product Type:	Primary Antibodies
Clone Name:	RB6-8C5
Applications:	FC, WB
Recommended Dilution:	Flow cytometry (1,2,3). Western blot (5).
Reactivity:	Mouse
Host:	Rat
Isotype:	IgG2b
Clonality:	Monoclonal
Specificity:	This antibody reacts with the Mouse myeloid differentiation antigen GR-1 (1,2).
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 Ig fraction Absorption emission: 495 nm / 528 nm
Concentration:	lot specific
Purification:	Protein G chromatography
Conjugation:	FITC
Storage:	Store the antibody at 2 - 8 °C for up to one month. For long term storage, aliquot and freeze unused portion at -20 °C in volumes appropriate for single usage. Avoid repeated freezing and thawing This product is photosensitive and should be protected from light.
Stability:	Shelf life: one year from despatch.
Gene Name:	lymphocyte antigen 6 complex, locus G
Database Link:	Entrez Gene 546644 Mouse P35461

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Background: GR-1 is a 25-30 kDa cell surface antigen and is expressed on myeloid cells but not lymphoid or erythroid cells. The expression of the Gr-1 antigen increases with granulocyte maturation (3) as shown by the distinct populations of bone-marrow cells this monoclonal antibody labels: negative, low positive and high positive. Expression is transient on cells of monocytic lineage (3).

Synonyms: Gr-1 Granulocyte marker

Note:

Protocol: 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 1 x 10e6 cells, representing 1 test).
4. To each tube, add 0.1 - 0.5 µg of antibody 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.

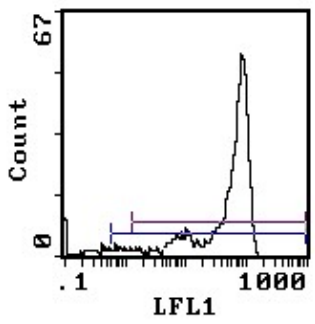
Media:
A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide (100 µl of 2M sodium azide in 100 mls).
B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 µl of 2M sodium azide in 100 mls).

Results:
Tissue Distribution by Flow Cytometry Analysis:
Mouse Strain: CBA/J
Cell Concentration : 1x10e6 cells per test
Antibody Concentration Used: 0.1 µg/10e6 cells
Isotypic Control: FITC Rat IgG2b
Cell Source Percentage of cells stained above control:
Thymus 1.5%
Whole Blood Monocytes 87.2%
Bone Marrow Macrophages 90.0%

(see picture below)

Strain Distribution by Flow Cytometry Analysis:
Procedure: see above
Cell Concentration: 1x10e6 cells per test
Antibody Concentration Used: 0.1 µg/10e6 cells
Strains Tested: BALB/c, C57BL/6, CBA, C3H/he, AKR
Positive: BALB/c, C57BL/6, CBA, C3H/he, AKR
Negative: none

Product images:



Cell Source: Bone Marrow Macrophages
Percentage of cells stained above control: 90.0%