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Product datasheet for CL046B

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

Product data:

Product Type:	Primary Antibodies
Clone Name:	RB6-8C5
Applications:	FC, IHC, WB
Recommended Dilution:	Flow Cytometry (Ref.1-3). Immunohistochemistry on Frozen and Paraffin Embedded Sections(Ref.6). Western blot (Ref.5). Complement-mediated depletion.
Reactivity:	Mouse
Host:	Rat
lsotype:	lgG2b
Clonality:	Monoclonal
Immunogen:	Mouse Granulocytes.
Specificity:	 Anti-Mouse Gr-1 monoclonal antibody reacts with the myeloid differentiation antigen Gr-1. (Ref.1,2). This 25-30 kDa cell surface antigen is expressed on myeloid cells but not lymphoid or erythroid cells. The expression of the Gr-1 antigen increases with granulocyte maturation (Ref.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 (Ref.3).
Formulation:	PBS containing 0.02% Sodium Azide as preservative and EIA grade BSA as a stabilizing protein to bring total protein concentration to 4-5 mg/ml Label: Biotin State: Liquid lg fraction
Concentration:	lot specific
Purification:	Protein G Chromatography
Conjugation:	Biotin
Storage:	Store undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer. Avoid repeated freezing and thawing.
Stability:	Shelf life: one year from despatch.



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	Ly6g Rat Monoclonal Antibody [Clone ID: RB6-8C5] – CL046B
Gene Name:	lymphocyte antigen 6 complex, locus G
Database Link:	<u>Entrez Gene 546644 Mouse</u> <u>P35461</u>
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 with cell separation medium.

2. Wash 2 times.

3. Resuspend the cells to a concentration of $2x10^{\circ}$ cells/ml in media A. Add 50 μ l of this

suspension to each tube (each tube will then contain 1×10^6 cells, representing 1 test).

4. To each tube, add 0.1-0.2 μg^{\star} of CL046B per 10 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 μl of secondary antibody FITC Streptavidin at 1/500 dilution.

9. Incubate tubes at 4°C for 30-60 minutes (It is recommended that tubes are protected from light since most fluorochromes are light sensitive).

10. Wash 2 times at 4°C.

11. Resuspend the cell pellet in 50 μl 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 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:

<u>Mouse Strain:</u> BALB/c <u>Cell Concentration:</u> 1x10⁶ cells per test <u>Antibody Concentration Used:</u> 0.1 µg/10⁶ cells <u>Isotypic Control:</u> Rat IgG2b

Cell: Source Percentage of cells stained above control:

Thymus: 1.5% Whole Blood: Granulocytes: 90.8%, Monocytes: 91.8% Bone Marrow: Granulocytes: 94.2%, Monocytes: 95.3%

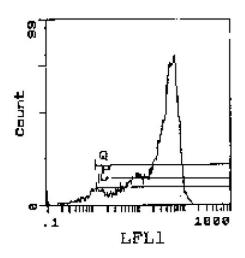
Cell Source: Bone Marrow Monocytes

Percentage of cells stained above control:95.3%

Strain Distribution by Flow Cytometry Analysis:

<u>Cell Concentration:</u> 1x10⁶ cells per test. <u>Antibody Concentration Used:</u> 0.1 µg/10⁶ cells. <u>Strains Tested:</u> BALB/c, C57BL/6, CBA, C3H/He. <u>Positive:</u> BALB/c, C57BL/6, CBA, C3H/He. <u>Negative:</u> none.

Product images:



Cell Source: Bone Marrow Monocytes. Percentage of cells stained above control: 95.3%

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