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Product datasheet for CL050B

Neutrophils Rat Monoclonal Antibody [Clone ID: 7/4]

Product data:

Product Type:	Primary Antibodies
Clone Name:	7/4
Applications:	FC
Recommended Dilution:	Flow Cytometry. (Reported to be useful in Immunohistochemistry on frozen and paraffin sections. Strains reported to be positive for the 7/4 clone are: AKR, C57BL/6, C57BL/10, C58, DBA/2, MF1, NZB, NZW, SJL, Swiss (PO) and 129J. Strains reported to be negative/weak for the 7/4 clone are: A2G, A/Sn, ASW, BALB/c, C3H/HEH and CBA.T6T6.)
Reactivity:	Mouse
Host:	Rat
Isotype:	IgG2a
Clonality:	Monoclonal
Immunogen:	Cultured bone marrow cells
Specificity:	This monoclonal antibody is specific for mouse neutrophils. It reacts with the 7/4 antigen that is a polymorphic 40 kD molecule expressed by polymorphonuclear cells, but absent on resident tissue macrophages.
Formulation:	PBS Label: Biotin State: Liquid purified Ig
Concentration:	lot specific
Purification:	Protein A affinity purified
Conjugation:	Biotin
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.
Stability:	Shelf life: one year from despatch.

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Note:

Protocol: **FLOW CYTOMETRY ANALYSIS:**

Method:

1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population using a NH4Cl lysing buffer.
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 1x10e6 cells, representing 1 test).
4. To each tube, add ~1.0 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.
7. Wash 2 times at 4°C.
8. Add 100 µl of detection reagent (Streptavidin-FITC) at 1:50 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.

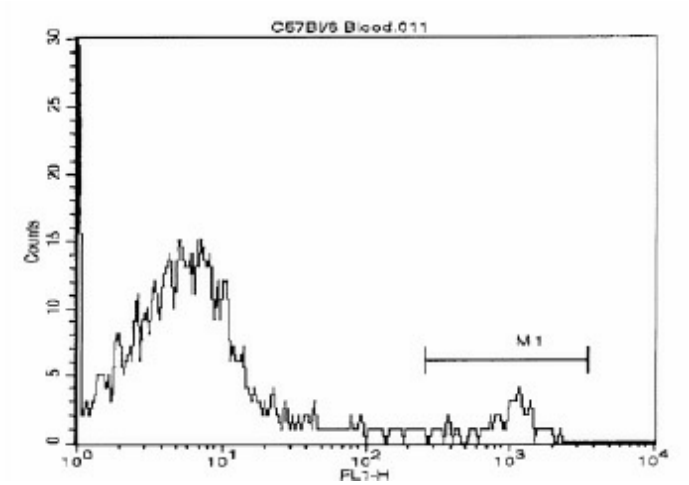
Media:

A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide (100 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: C57BL/6
Cell Concentration: 1x10e6 cells per test
Antibody Concentration Used: 1.0 ug/10e6 cells
Isotypic Control: Biotin Rat IgG2a

Product images:



Cell Source: Blood - Percentage of cells stained above control: 7.6%