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Monoclonal Antibody to Neutrophils - PE

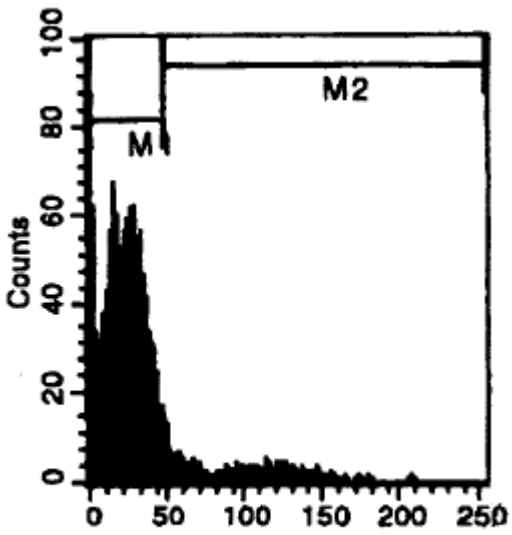
Catalog No.:	CL050R
Quantity:	50 µg
Concentration:	0.1 mg/ml
Host / Isotype:	Rat / IgG2a
Clone:	7/4
Immunogen:	Cultured bone marrow cells
Format:	**State:** Liquid purified Ig fraction. **Purification:** Affinity Chromatography on Protein A. **Buffer System:** PBS with 0.09% Sodium Azide as preservative and 1% EIA grade BSA as a stabilizing protein. **Label:** PE – conjugated *Absorption / Emission:* 488 nm / 575 nm
Applications:	Flow Cytometry (See ''Protocol''). Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Specificity:	Reacts with Mouse Neutrophils. Strains to be positive for the 7/4 clone are: AKR, C57BL/6, C57BL/10, C58, DBA/2, MF1, NZB, NZW, SJL, Swiss (PO), 129J. Strains reported to be negative/weak for the 7/4 clone: A2G, A/Sn, ASW, BALB/2, C3H/HEH and CBA.T6T6.
Species Reactivity:	**Tested:** Mouse.
Add. Information:	This clone has also been reported to work in Western blotting and immunohistochemistry (frozen and paraffin sections, 2).
Storage:	Store the antibody undiluted at 2-8°C. **DO NOT FREEZE.** This product is photosensitive and should be protected from light. Shelf life: one year from despatch.
General References:	1. Hirch, S. and Gordon, S. (1983). Polymorphic expression of a neutrophil differentiation antigen revealed by a monoclonal antibody 7/4. Immunogenetics 18:229-239. 2. Gordon, S. et al. (1992). Antigen markers of macrophage differentiation in murine tissues. Curr. Top. Microbio. Immunol. 181:1-37.
Protocols:	**FLOW CYTOMETRY ANALYSIS:** **Method:** 1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population using an 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 CL050R 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: C57BL/6
Cell Concentration: 1x10e6 cells per test
Antibody Concentration Used: 1.0 µg/10e6 cells
Isotypic Control: PE Rat IgG2a

Pictures:



LFL2

Cell Source: Peripheral Blood Leukocytes
- Percentage of cells stained above control: 11.4%