

Monoclonal Antibody to Neutrophils - PE

Catalog No.: CL050RX

Quantity: 0.3 mg

Concentration: 0.1 mg/ml

Host / Isotype: Rat / IgG2a

Clone: 7/4

Immunogen: Cultured bone marrow cells

Format: State: Liquid

Purification: Protein A affinity purified Ig

Buffer System: PBS with 0.09% NaN3 as preservative and EIA grade BSA as a stabilizing

protein to bring total protein concentration to 4-5 mg/ml

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:** Mouse.

Other species not tested.

Add. Information: This clone has also been reported to work in Western blotting and immunohistochemistry

(frozen and paraffin sections, 2).

Store at 4°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.

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Material Safety Datasheets are available at www.acris-antibodies.com or on request.



- 3. Resuspend the cells to a concentration of 2x10e7 cells/ml in media A. Add 50 μ l of this suspension to each tube (each tube will then contain 1x10e6 cells, representing 1 test).
- 4. To each tube, add \sim 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 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).

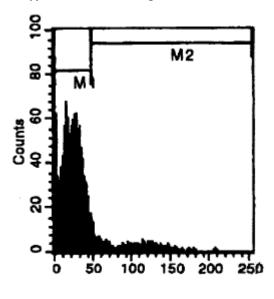
Results - Tissue Distribution:

Mouse Strain: C57BL/6

<u>Cell Concentration</u>: 1x10e6 cells per test <u>Antibody Concentration Used</u>: 1.0 μg/10e6 cells

Isotypic Control: PE Rat IgG2a

Pictures:



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

control: 11.4%

LFL2