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Product datasheet for CL050FX

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

Product data:

Product Type:	Primary Antibodies
Clone Name:	7/4
Applications:	FC
Recommended Dilution:	This antibody is suitable for use in Flow cytometry. This clone has also been reported to be useful for Immunohistochemistry (both frozen and paraffin sections).
Reactivity:	Mouse
Host:	Rat
Isotype:	IgG2a
Clonality:	Monoclonal
Immunogen:	Cultured bone marrow cells.
Specificity:	This antibody is specific for detecting mouse neutrophils. 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.
Formulation:	PBS with 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: FITC State: Liquid Ig fraction.
Concentration:	lot specific
Conjugation:	FITC
Storage:	Store the antibody at 2-8°C for one month or (in aliquots) at -20°C for longer. Avoid prolonged exposure to light. Avoid freeze/thaw cycles.
Stability:	Shelf life: one year from despatch.

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Note:

Actual results of Flow cytometric analysis
Mouse strain: C57BL/6
Cell source: Peripheral Blood Leukocytes
Cell concentration: 1x10e6 cells per test
Antibody concentration: 1.0 µg/10e6 cells
Isotypic control: FITC Rat IgG2a
Percentage of cells stained above control: 21.84%

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 1 x 10e6 cells, representing 1 test).
4. To each tube, add ~1.0 µg of CL050F 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).