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Product datasheet for CL030RX

Itga4 Rat Monoclonal Antibody [Clone ID: R1-2]

Product data:

Product Type:	Primary Antibodies
Clone Name:	R1-2
Applications:	FC
Recommended Dilution:	**Flow Cytometry** (See Protocols). This clone is also reportded to work in**Immunoprecipitation** and **Immunohistochemistry**. (1,2,3)
Reactivity:	Mouse
Host:	Rat
Isotype:	IgG2b
Clonality:	Monoclonal
Immunogen:	Peyers Patch HEV binding lymphoma line (TK1). Donor: Fisher Spleen. Fusion Partner: P3x63Ag8.653.
Specificity:	This CD49d Monoclonal Antibody reacts with Alpha-4 integrin (CD49d/ITGA4).
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: PE State: Liquid purified Ig fraction
Concentration:	lot specific
Purification:	Protein G Chromatography
Conjugation:	PE
Storage:	Store the antibody undiluted at 2-8°C. **DO NOT FREEZE!** This product is photosensitive and should be protected from light.
Stability:	Shelf life: one year from despatch.
Gene Name:	integrin alpha 4
Database Link:	Entrez Gene 16401 Mouse Q00651

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Background: CD49d helps to mediate cell-cell and cell-matrix interactions. Alpha-4 integrin combines with b1 and b7 integrin to form VLA-4 and LPAM-1 (Peyers patch homing receptor) respectively. VLA-4 is expressed on most peripheral lymphocytes, thymocytes and monocytes. LPAM-1 is found on peripheral lymphocytes, but few thymocytes. Fibronectin and VCAM-1 act as ligands for both VLA-4 and LPAM-1. LPAM-1 also binds the mucosal vascular addressin MAdCAM-1. (1)

Synonyms: Integrin alpha-4, Integrin alpha-IV, VLA-4, VLA4

Note: Protocol: **Flow Cytometry Analysis:**

Method:

1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population with Lympholyte®-M cell separation medium.
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 1x106 cells, representing 1 test).
4. To each tube, add ~1.0 µg* of CL030R or CL030RX.
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:

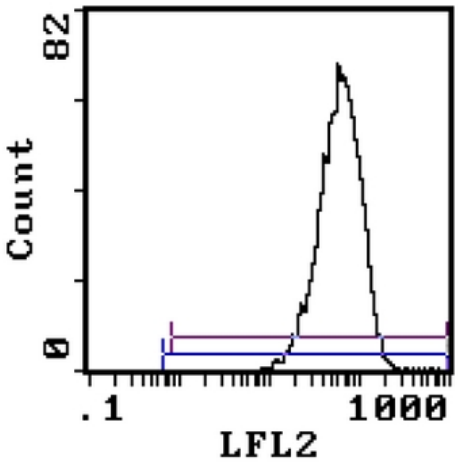
Tissue Distribution by Flow Cytometry Analysis:
Mouse Strain: BALB/c
Cell Concentration : 1x10e6 cells per test
Antibody Concentration Used: 1.0 µg/106 cells
Isotypic Control: PE Rat IgG2b

Cell Source Percentage of cells stained above control:
TK1 cell line: 100%
Thymus: 93.5%
Spleen: 92.7%

Bone Marrow 88.0%

Strain Distibution:
Cell Concentration: 1x10e6 cells per test
Antibody Concentration Used: 1.0 µg/10e6 cells
Strains Tested: BALB/c, C57BL/6, C3H/He, CBA/J, AKR
Positive: BALB/c, C57BL/6, C3H/He, CBA/J, AKR
Negative: None.

Product images:



Cell Source: TK1 cell line. Percentage of cells stained above control: 100%