

Monoclonal Antibody to MHC Class II I-Ad - PE

Catalog No.:	CL090R
Quantity:	50 µg
Concentration:	0.1 mg/ml
Host / Isotype:	Mouse / IgG2a
Clone:	34-5-3S
Immunogen:	BDF splenocytes Remarks: R-Phycoerythrin conjugates are produced under license and protected under Stanford University held patents 4,520,110; 4,542,104; 4,859,582; 5,055,556 (U.S.): 76695 (EPC): 548440 (Australia): 1,179,942 (Canada): and 1,594,827 (Japan).
Format:	State: Liquid Ig raction Purification: Protein G affinity chromatography Buffer System: 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: PE – R-Phycoerythrin
Applications:	Flow cytometry. Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Specificity:	This antibody is a cytotoxic monoclonal antibody specific for cells expressing the Ia antigen coded for by the A subregion of the d, b, p, and q haplotypes (ie. I-Ad,b,p,q). Results of flow cytometric analysis (Tissue distribution): Mouse Strain: BALB/c Cell concentration : 1x10e6 cells per test Antibody concentration used: 0.1 µg/10e6 cells Isotypic control: PE Mouse IgG2a Cell source percentage of cells stained above control: Spleen 52.0% (see picture below) Lymph Node 13.5% (Strain distribution): Antibody concentration: 0.2 µg/10e6 cells Strains Tested: A.TH, A.TL, C3H/He, C57BL/6, DBA/1 Positive: C57BL/6, DBA/1 Negative: A.TH, A.TL, C3H/He
Storage:	Store at 2-8°C. DO NOT FREEZE. Avoid prolonged exposure to light. Shelf life: one year from despatch.

For research and in vitro use only. Not for diagnostic or therapeutic work.

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

Antibody Hotline - Technical Questions - Antibody Location Service
Free Call: 0800-2274746 (Germany only) - www.acris-antibodies.com

- General References:**
1. Ozato, K. et al. 1982. Monoclonal Antibodies to Mouse Major Histocompatibility Complex Antigens. *Transplantation*. 34: 113-120.
 2. Ahn, H.J. et al. 1997. A Mechanism Underlying Synergy Between IL-12 and IFN-g-Inducing Factor in Enhanced Production of IFN-g. *Journal of Immunology*. 159: 2125-2131.

Protocols:

FLOW CYTOMETRY ANALYSIS:

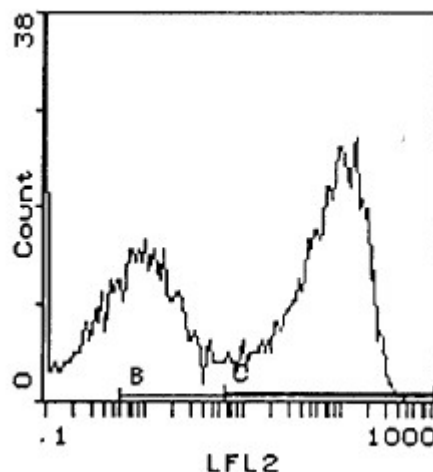
Method:

1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population.
2. Wash 2 times.
3. Resuspend the cells to a concentration of 2×10^7 cells/ml in media A. Add 50 μ l of this suspension to each tube (each tube will then contain 1×10^6 cells, representing 1 test).
4. To each tube, add 0.2 - 0.1 μ g of CL090R per 10^6 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 μ l 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).

Pictures:



Flow cytometric analysis: Cell source is spleen. Percentage of cells stained above control: 52.0%

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