

Product datasheet for **AM08081FC-S**

MHC Class I H2 Kd/Dd Mouse Monoclonal Antibody [Clone ID: 34-1-2S]

Product data:

Product Type:	Primary Antibodies
Clone Name:	34-1-2S
Applications:	FC
Recommended Dilution:	Flow Cytometry.
Reactivity:	Mouse
Host:	Mouse
Isotype:	IgG2a
Clonality:	Monoclonal
Immunogen:	BDF splenocytes <u>Donor:</u> C3H spleen <u>Fusion Partner:</u> Sp2/0-Ag14
Specificity:	Anti H-2KdDd mAb reacts with both H-2Kd and H-2Dd products. The antibody also cross reacts with Kbsrpq. This K-D cross reaction indicates the presence of shared specificities between the two separate H-2 regions.
Formulation:	PBS containing 0.02% sodium azide (NaN ₃) as preservative and EIA grade BSA as a stabilizing protein Label: FITC State: Liquid purified Ig fraction Label: Fluorescein isothiocyanate isomer 1
Concentration:	lot specific
Purification:	Affinity chromatography on Protein G
Conjugation:	FITC
Storage:	Store the antibody undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer. Avoid repeated freezing and thawing.
Stability:	Shelf life: one year from despatch.



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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 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 0.5 - 0.2 μ g of this antibody 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).

Results:

Tissue Distribution by Flow Cytometry Analysis:

Mouse Strain: BALB/c

Cell Concentration: 1×10^6 cells per test

Antibody Concentration Used: 0.5 μ g/ 10^6 cells

Isotypic Control: FITC Mouse IgG2a

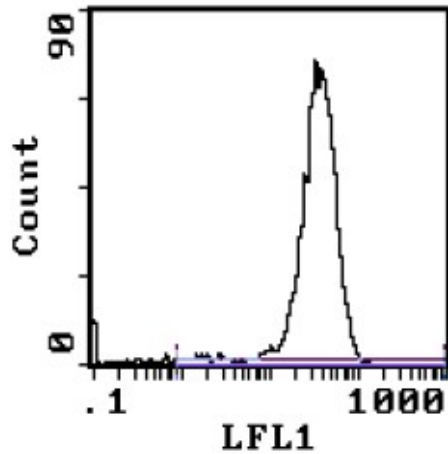
Percentage of cells stained above control:

Thymus 91.9%

Spleen 95.1%

Lymph Node 100%

Product images:



Cell Source: Spleen Percentage of cells stained above control: 95.1%

Strain Distribution by Flow Cytometry Analysis:

Procedure: As above

Antibody Concentration: 0.2 µg/10⁶ cells

Strains Tested:

Strain	H-2 Loci Alleles								±/-
	<u>K</u>	<u>A_u</u>	<u>A_β</u>	<u>E_β</u>	<u>E_α</u>	<u>C4</u>	<u>C4S</u>	<u>D</u>	
C3H/He	k	k	k	k	k	k	k	k	-
C57BL/6	b	b	b	b	b	b	b	b	(+/-)
BALB/c	d	d	d	d	d	d	d	d	+
DBA/1	q	q	q	q	q	q	q	q	+
SJL	s	s	s	s	s	s	s	s	+
B10.M	f	f	f	f	f	f	f	f	(+/-)
A.TH	s	s	s	s	s	s	s	d	+
A.TL	s	k	k	k	k	k	k	d	+
B10.A(3R)	b	b	b	b/k	k	d	d	d	+
P/J	p	p	p	p	p	p	p	p	(+/-)

For a more detailed strain distribution - see reference 1.