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Product datasheet for AM31864BT-N

Cd55 Hamster Monoclonal Antibody [Clone ID: RIKO-3]

Product data:

Product Type:	Primary Antibodies
Clone Name:	RIKO-3
Applications:	ELISA, FC, WB
Recommended Dilution:	**ELISA.** **Western Blot.** **Flow Cytometry** (See Protocols).
Reactivity:	Mouse
Host:	Hamster
Isotype:	IgG
Clonality:	Monoclonal
Immunogen:	GPI-DAF transfected CHO cells from Armenian Hamster spleen. Fusion Partner: P3U1
Specificity:	This DAF Monoclonal Antibody (Clone: RIKO-3) detects Mouse DAF. Other species not tested.
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: Biotin State: Liquid purified IgG fraction.
Concentration:	lot specific
Purification:	Protein G Affinity Chromatography.
Conjugation:	Biotin
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.
Gene Name:	CD55 molecule, decay accelerating factor for complement
Database Link:	Entrez Gene 13136 Mouse Q61475

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Background: DAF is one of several membrane proteins that prevent host cells from homologous complement attack. The two mouse DAF isoforms are effective against rat complement, but not against human and guinea pig complement. GPI-DAF is expressed on erythrocytes, spleen and testis while TM-DAF (transmembrane-DAF) is expressed only in testis.
Both types of DAF transfectants avoided C3 deposition more successfully than non-transfectant cells.

Synonyms: CR; CROM; DAF; TC

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

Method:

1. Prepare 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 1x10e6 cells, representing 1 test).
4. To each tube, add 1.0 µg of AM31864BT-N per 1x10e6 cells.
5. Vortex the tubes to ensure thorough mixing of antibody and cells.
6. Incubate the tubes for 30 minutes at 4°C.
7. Wash 2 times at 4°C.
8. Add 100 µl of secondary antibody (Streptavidin-FITC) at a 1/500 dilution.
9. Incubate the tubes at 4°C for 30-60 minutes.
(It is recommended that the tubes are protected from light since most fluorochromes are light sensitive).
10. Wash 2 times at 4°C in media B.
11. Resuspend the cell pellet in 50 µl ice cold media B.
12. 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.

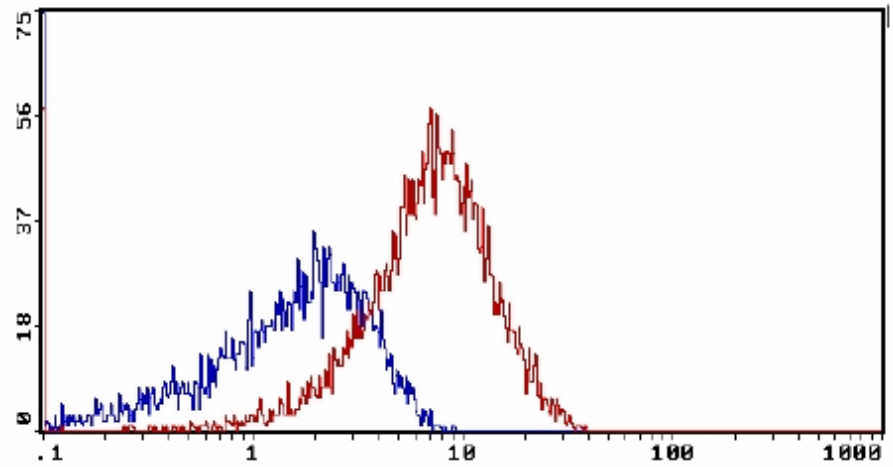
Media:

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:

Mouse Strain: Balb/c.
Cell Concentration : 1x10e6 cells per test.
Antibody Concentration Used: 0.2 µg/10e6 cells.
Isotypic Control: Armenian Hamster IgG-Biotin.

Product images:



Cell Source: Red Blood Cells. Percentage of cells stained above control: 47.9%