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Product datasheet for AM20011AF-N

Integrin alpha-7 / ITGA7 Mouse Monoclonal Antibody [Clone ID: 3C12]

Product data:

Product Type:	Primary Antibodies
Clone Name:	3C12
Applications:	FC, IF
Recommended Dilution:	**Flow Cytometry:** 10-20 µg/ml (final concentration). **Immunocytochemistry:** 10 µg/ml. *Positive Control:* C2C12. Detailed Procedure is provided in the following**Protocols.**
Reactivity:	Mouse
Host:	Mouse
Isotype:	IgG1
Clonality:	Monoclonal
Immunogen:	Mouse myoblasts.
Specificity:	This antibody reacts with Mouse Integrin alpha-7.
Formulation:	PBS, pH 7.2 containing 50% Glycerol without preservatives. State: Azide Free State: Liquid purified IgG fraction.
Concentration:	lot specific
Purification:	Protein-A Agarose Chromatography.
Conjugation:	Unconjugated
Storage:	Upon receipt, store undiluted (in aliquots) at -20°C. Avoid repeated freezing and thawing.
Stability:	Shelf life: one year from despatch.
Database Link:	Q61738

View online »

Background:

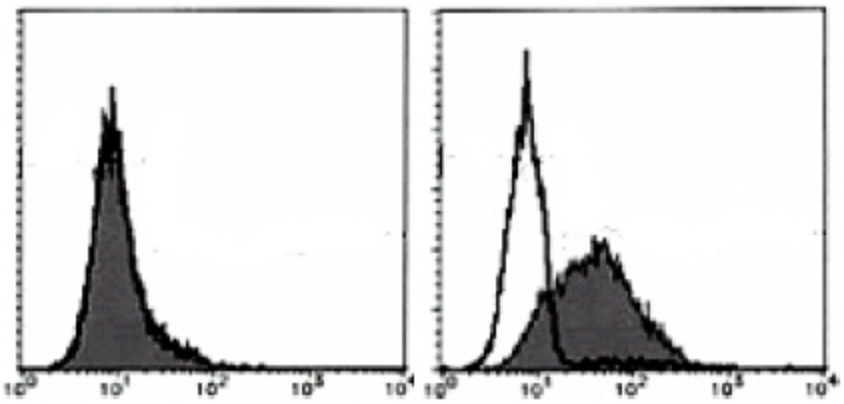
The integrin family of adhesion molecules participate in important cell-cell and cell-extracellular matrix interactions in a diverse range of biological processes. Integrins are heterodimers consisting of an alpha subunit and a beta subunit. Both alpha and beta subunits are transmembrane proteins with large extracellular domains (>100 kDa for alpha subunit and >75 kDa for beta subunit) that interact with extracellular matrix proteins and relatively small cytoplasmic domains (50 amino acids or less, except for the beta-4 subunit) that interact with cytoskeletal proteins. The adhesiveness of integrins is dynamically regulated in response to cytoplasmic signals, termed “inside-out” signaling. It has been reported that, upon ligand binding, integrins regulate many intracellular signaling pathways that involve cytoplasmic alkalization, intracellular Ca^{2+} fluctuation, inositol lipid metabolism, protein kinase C, MAP kinase and phosphatidyl inositol kinase. Integrin alpha-7 is a specific cellular receptor for the basement membrane protein laminin-1, as well as for the laminin isoforms-2 and -4. The alpha-7 subunit is expressed mainly in skeletal and cardiac muscle and may be involved in differentiation and migration processes during myogenesis. Absence of integrin alpha-7 results in muscular dystrophy is revealed.

Note: This product was originally produced by MBL International.

Protocol: **Flow Cytometric Analysis for Adherent Cells**
We usually use Fisher tubes or equivalents as reaction tubes for all steps after 2).
1) Detach the cells from the culture dish by using cell dissociation buffer
2) Wash the cells 3 times with washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.1% NaN_3].
3) Resuspend the cells with washing buffer ($5x10^6$ cells/ml).
4) Add 50 µl of the cell suspension into each tube, and centrifuge at 500 x g for 1 minute at RT (20~25°C). Remove supernatant by careful aspiration.
5) Add 10 µl of normal goat serum containing 1 mg/ml normal human IgG and 0.1% NaN_3 to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
6) Add 40 µl of the primary antibody at the concentration suggested in the APPLICATIONS, diluted in the washing buffer. Mix well and incubate for 30 minutes at RT.
7) Add 1 mL of the washing buffer followed by centrifugation at 500 x g for 1 minute at RT. Remove supernatant by careful aspiration.
8) Add 30 µl of 1/100 FITC conjugated anti-Mouse IgG diluted with the washing buffer. Mix well and incubate for 15 minutes at RT
9) Add 1 ml of the washing buffer followed by centrifugation at 500 x g for 1 minute at RT. Remove supernatant by careful aspiration.
10) Resuspend the cells with 500 µl of the washing buffer and analyze by a flow cytometer.
Positive Control for Flow Cytometry: C2C12

Immunocytochemistry
1) Add the primary antibody diluted with PBS as suggest in the APPLICATIONS onto the cells and incubate for 1 hour at RT. (Optimization of antibody concentration or incubation condition are recommended if necessary.)
2) Prepare a wash container such as a 500 mL beaker with a magnetic stirrer. Then wash the cultured cells on the glass slide by soaking the slide with plenty of PBS in the wash container for 5 minutes. Take care not to touch the cells. Repeat wash once more.
3) Add 30 µl of 1/40 FITC conjugated anti-mouse IgG diluted with PBS onto the cells. Incubate for 30 minutes at RT. Keep out light by covering with aluminum foil.
4) Wash the slide in plenty of PBS as in step 2).
5) Wipe excess liquid from slide but take care not to touch the cells. Never leave the cells to dry.
6) Promptly add PermafluorTM aqueous mounting medium onto the slide, then put a cover slip on it.

Product images:



Flow Cytometric analysis of mouse Integrin alpha-7 expression on NIH/3T3 (Left) and C2C12 (Right). Open Histogram indicates the reaction of Isotypic Control to the cells. Shaded Histograms indicate the reaction of ITGA7 antibody to the cells.