

Anti-ICAM1 Antibody Picoband™ (monoclonal, 6F2C3)

Catalog # ABO16259

Product Information

Application	WB, IHC, FC
Primary Accession	P05362
Host	Mouse
Isotype	Mouse IgG1
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-ICAM1 Antibody Picoband™ (monoclonal, 6F2C3) . Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.

Additional Information

Gene ID	3383
Other Names	Intercellular adhesion molecule 1, ICAM-1, Major group rhinovirus receptor, CD54, ICAM1
Calculated MW	57825
Application Details	Western blot, 0.25-0.5 µg/ml, Human Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human Flow Cytometry, 1-3 µg/1x10 ⁶ cells, Human
Source	Eukaryota
Contents	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na ₂ HPO ₄ .
Clone Names	Clone: 6F2C3
Immunogen	E. coli-derived human ICAM1 recombinant protein (Position: Q28-R268).
Purification	Immunogen affinity purified.
Storage	At -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freezing and thawing.

Protein Information

Name	ICAM1 (HGNC:5344)
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Function

Cell adhesion molecule that functions as a receptor ligand of the signaling receptor ITGAL:ITGB2/LFA-1 (lymphocyte-function associated (LFA) molecule 1) ensuring leukocyte cell-cell adhesion, by providing a calibrated system to namely adjust T-cell killing to the antigen stimulation strength (PubMed:[3086451](#), PubMed:[3340213](#), PubMed:[38195629](#)). Also functions as a ligand receptor of the signaling receptor ITGAM:ITGB2/MAC-1 ensuring adhesion between stimulated neutrophils and stimulated endothelial cells (PubMed:[1980124](#)). During leukocyte trans-endothelial migration, ICAM1 engagement promotes the assembly of endothelial apical cups through ARHGEF26/SGEF and RHOG activation (PubMed:[17875742](#)). Promotes cell aggregation in epithelial cells through interaction with MUC1 (PubMed:[11173916](#)).

Cellular Location

Cell membrane; Single-pass type I membrane protein

Tissue Location

Expressed on non-hematopoietic cells such as vascular endothelial cells, thymic epithelial cells, certain other epithelial cells, and fibroblasts, and on hematopoietic cells such as tissue macrophages, mitogen-stimulated T lymphocyte blasts, and germinal center dendritic cells in tonsils, lymph nodes, and Peyer's patches (PubMed:[3086451](#)). Expressed in low amounts on peripheral blood leukocytes (PubMed:[3086451](#)).

Background

CD54, also known as ICAM-1. Intercellular adhesion molecule-1 (ICAM1) is a ligand for lymphocyte function-associated (LFA) antigens. ICAM-1 is an integral membrane protein, a member of the immunoglobulin superfamily, and a ligand for LFA-1, a beta 2 leukocyte integrin. This protein is the major human rhinovirus receptor. The ICAM1 gene is mapped to human chromosome 19. In humans, lymphocyte adhesion to cells is mediated by the protein heterodimer CD11a/CD18 (Leu-CAMa, LFA-1) and its ligand CD54 (ICAM-1).

Images

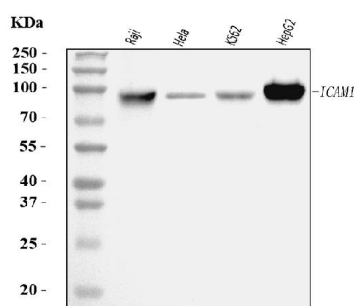


Figure 1. Western blot analysis of ICAM1 using anti-ICAM1 antibody (M00171-3).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human Raji whole cell lysates,
Lane 2: human Hela whole cell lysates,
Lane 3: human K562 whole cell lysates,
Lane 4: human HepG2 whole cell lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-ICAM1 antigen affinity purified monoclonal antibody (Catalog # M00171-3) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for ICAM1 at approximately 90-110 kDa. The expected band size for ICAM1 is at 59 kDa.

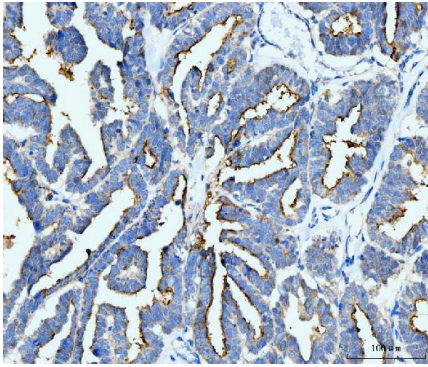


Figure 2. IHC analysis of ICAM1 using anti-ICAM1 antibody (M00171-3).

ICAM1 was detected in a paraffin-embedded section of human ovarian serous adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-ICAM1 Antibody (M00171-3) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

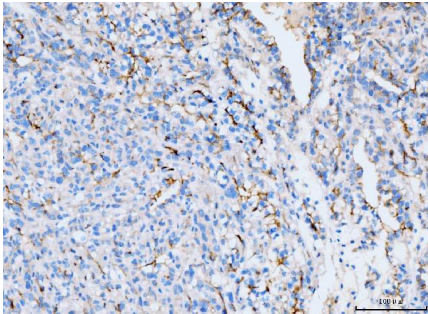


Figure 3. IHC analysis of ICAM1 using anti-ICAM1 antibody (M00171-3).

ICAM1 was detected in a paraffin-embedded section of human renal carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-ICAM1 Antibody (M00171-3) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

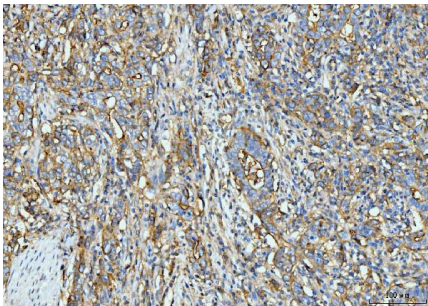


Figure 4. IHC analysis of ICAM1 using anti-ICAM1 antibody (M00171-3).

ICAM1 was detected in a paraffin-embedded section of human rectal moderately differentiated adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-ICAM1 Antibody (M00171-3) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

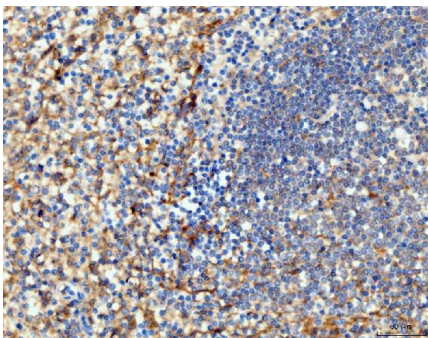
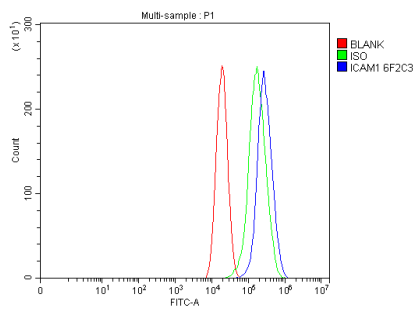


Figure 5. IHC analysis of ICAM1 using anti-ICAM1 antibody (M00171-3).

ICAM1 was detected in a paraffin-embedded section of human spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-ICAM1 Antibody (M00171-3) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

Figure 6. Flow Cytometry analysis of Caco-2 cells using anti-ICAM1 antibody (M00171-3).



Overlay histogram showing Caco-2 cells stained with M00171-3 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-ICAM1 Antibody (M00171-3, $1 \mu\text{g}/1 \times 10^6$ cells) for 30 min at 20°C . DyLight®488 conjugated goat anti-mouse IgG (BA1126, $5\text{-}10 \mu\text{g}/1 \times 10^6$ cells) was used as secondary antibody for 30 minutes at 20°C . Isotype control antibody (Green line) was mouse IgG ($1 \mu\text{g}/1 \times 10^6$) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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