Anti-ICAM1 Antibody [F11-A4]

M1505-7



Product Type:	Mouse monoclonal IgG2a/IgG2c, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, FC
Molecular Wt:	Predicted band size: 58 kDa
Clone number:	F11-A4
Description:	ICAM-1 is a member of the immunoglobulin superfamily, the superfamily of proteins including antibodies and T-cell receptors. The structure of ICAM-1 is characterized by heavy glycosylation, and the protein's extracellular domain is composed of multiple loops created by disulfide bridges within the protein. ICAM-1 can be induced by interleukin-1 (IL-1) and tumor necrosis factor (TNF) and is expressed by the vascular endothelium, macrophages, and lymphocytes. ICAM-1 is a ligand for LFA-1 (integrin), a receptor found on leukocytes. More recently, ICAM-1 has been characterized as a site for the cellular entry of human rhinovirus. ICAM-1 and soluble ICAM-1 have antagonistic effects on the tight junctions forming the blood-testis barrier, thus playing a major role in spermatogenesis. ICAM-1 has been implicated in subarachnoid hemorrhage (SAH). Levels of ICAM-1 are shown to be significantly elevated in patients with SAH over control subjects in many studies.
lmmunogen:	Synthetic peptide within Human ICAM1 aa 483-532 / 532.
Positive control:	Ramos cell lysate, Raji cell lysate, HUVEC cell lysate, Raji.
Subcellular location:	Membrane.
Database links:	SwissProt: P05362 Human
Recommended Dilutions: WB IF-Cell FC	1:1,000 1:100 1:1,000
Storage Buffer:	1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!C$ after thawing. Aliquot store at -20 $^\circ\!C$ or -80 $^\circ\!C$. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

Fig1: Western blot analysis of ICAM1 on different lysates with Mouse anti-ICAM1 antibody (M1505-7) at 1/1,000 dilution.

Lane 1: Ramos cell lysate Lane 2: Raji cell lysate Lane 3: HUVEC cell lysate

Lysates/proteins at 30 µg/Lane.

Predicted band size: 58 kDa Observed band size: 80 kDa

Exposure time: 2 minutes 30 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (M1505-7) at 1/1,000 dilution was used in 5% NFDM/TBST at 4° C overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

Fig2: Western blot analysis of ICAM1 on different lysates with Mouse anti-ICAM1 antibody (M1505-7) at 1/1,000 dilution.

Lane 1: HAP1-parental cell lysate Lane 2: HAP1-ICAM1 KD cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 58 kDa Observed band size: 80 kDa

Exposure time: 15 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (M1505-7) at 1/1,000 dilution was used in K1803 at 4° C overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.



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35 - GAPDH

HAP1 WT KD

ICAM1

kDa 250 -150 -

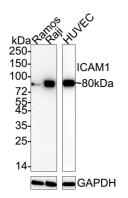
100

75

55 45

35 25

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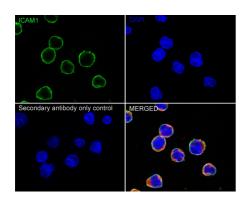


Fig3: Immunocytochemistry analysis of Raji cells labeling ICAM1 with Mouse anti-ICAM1 antibody (M1505-7) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-ICAM1 antibody (M1505-7) at 1/100 dilution in 1% BSA in PBST overnight at 4 $^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluorTM 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor TM 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

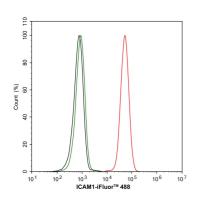


Fig4: Flow cytometric analysis of Raji cells labeling ICAM1.

Cells were fixed and permeabilized. Then stained with the primary antibody (M1505-7, 1/1,000) (red) compared with Mouse IgG Isotype Control (green). After incubation of the primary antibody at +4 °C for an hour, the cells were stained with a iFluor TM 488 conjugate-Goat anti-Mouse IgG Secondary antibody (HA1125) at 1/1,000 dilution for 30 minutes at +4 °C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

- "Interaction of coxsackievirus A21 with its cellular receptor, ICAM-1." Xiao C., Bator C.M., Bowman V.D., Rieder E., He Y., Hebert B., Bella J., Baker T.S., Wimmer E., Kuhn R.J., Rossmann M.G. J. Virol. 75:2444-2451(2001)
- "MARCH-IX mediates ubiquitination and downregulation of ICAM-1." Hoer S., Smith L., Lehner P.J. FEBS Lett. 581:45-51(2007)
- "RhoG regulates endothelial apical cup assembly downstream from ICAM1 engagement and is involved in leukocyte trans-endothelial migration." van Buul J.D., Allingham M.J., Samson T., Meller J., Boulter E., Garcia-Mata R., Burridge K. J. Cell Biol. 178:1279-1293(2007)

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