

Anti-ICAM1 Antibody [PSH17-38] - BSA and Azide free

HA751626



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Mouse
Applications:	IHC-Fr, IHC-P, WB, IF-Cell, FC, IP, IF-Tissue
Molecular Wt:	Predicted band size: 60 kDa
Clone number:	PSH17-38

Description: Cell adhesion molecules (CAMs) are a family of closely related cell surface glycoproteins involved in cell-cell interactions during growth and are thought to play important, yet separate, roles in embryogenesis and development. The intracellular adhesion molecule-1 (ICAM-1), also referred to as CD54, is an integral membrane protein of the immunoglobulin superfamily and recognizes the beta2alpha1 and beta2alphaM Integrins. ICAM-2 functions as a ligand for lymphocyte function-associated antigen-1 (LFA-1) and is involved in leukocyte adhesion. ICAM-3 is highly expressed on the surface of human eosinophils and, when bound to ligand, may inhibit eosinophil inflammatory responses and survival. ICAM-4, also known as LW glycoprotein, interacts with Integrins alphaLbeta2, alphaMbeta2, alpha4beta1, the alphaV family and alphaIIbbeta3, and selective binding to different integrins may be relevant to the pathology in a number of red blood cell associated diseases. Lastly, ICAM-5, expressed on telencephalic neurons, binds CD11a/CD18 and thus may act as an adhesion molecule for leukocyte binding in the central nervous system.

Immunogen: Recombinant protein within mouse ICAM1 aa 1-485.

Positive control: Mouse lung tissue, RAW264.7 cell lysate, Mouse spleen tissue lysate, Mouse kidney tissue lysate, RAW264.7, mouse spleen cells.

Subcellular location: Membrane.

Database links: SwissProt: P13597 Mouse

Storage Buffer: 1*PBS (pH7.4).

Storage Instruction: Store at 2-8°C. Avoid freeze.

Purity: Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

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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

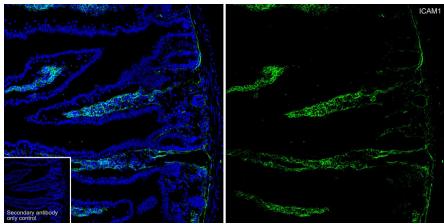


Fig1: This data was developed using HA723921, the same antibody clone in a different buffer formulation.

Application: IHC-Fr

Species: Mouse

Site: small intestine

Sample: Frozen section

Antibody concentration: 1/1,000

Antigen retrieval: Not required

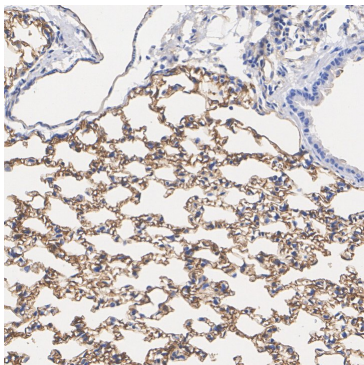


Fig2: This data was developed using HA723921, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded mouse lung tissue with Rabbit anti-ICAM1 antibody (HA723921) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA723921) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

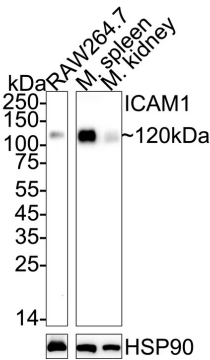
Fig3: This data was developed using HA723921, the same antibody clone in a different buffer formulation.

Western blot analysis of ICAM1 on different lysates with Rabbit anti-ICAM1 antibody (HA723921) at 1/10,000 dilution.

Lane 1: RAW264.7 cell lysate (20 µg/Lane)

Lane 2: Mouse spleen tissue lysate (30 µg/Lane)

Lane 3: Mouse kidney tissue lysate (30 µg/Lane)



Predicted band size: 60 kDa

Observed band size: 120 kDa

Exposure time: 3 minutes; 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 (HA723921) at 1/10,000 dilution was used in primary antibody dilution (K1803) at 4 °C overnight. Goat Anti-Rabbit IgG -

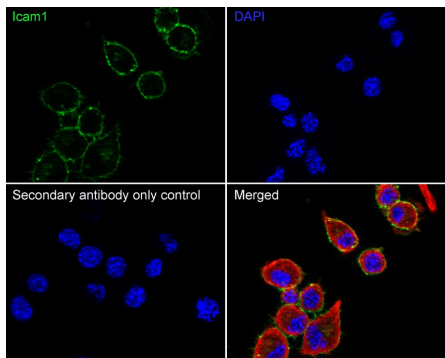


Fig4: This data was developed using HA723921, the same antibody clone in a different buffer formulation.

Immunocytochemistry analysis of RAW264.7 cells labeling ICAM1 with Rabbit anti-ICAM1 antibody (HA723921) at 1/200 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 Rabbit anti-ICAM1 antibody (HA723921) at 1/200 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) 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 (HA601187, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

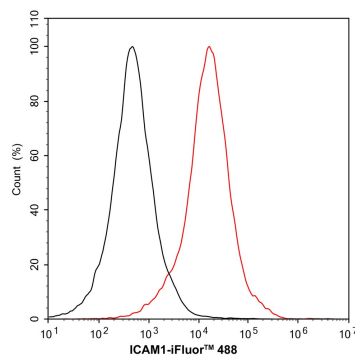


Fig5: This data was developed using HA723921, the same antibody clone in a different buffer formulation.

Flow cytometric analysis of mouse spleen cells labeling ICAM1.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA723921, 1/2,000) (red). After incubation of the primary antibody at +4°C for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) 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).

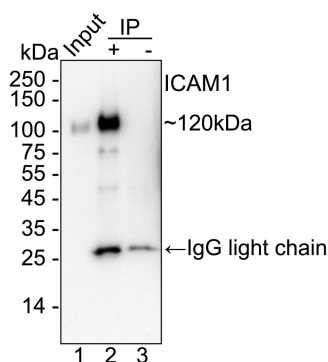


Fig6: This data was developed using HA723921, the same antibody clone in a different buffer formulation.

ICAM1 was immunoprecipitated from 0.2 mg mouse spleen tissue lysate with HA723921 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using HA723921 at 1/10,000 dilution. HRP Conjugated Anti-Rabbit IgG for IP Nano-secondary antibody at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: Mouse spleen tissue lysate (input)

Lane 2: HA723921 IP in mouse spleen tissue lysate

Lane 3: Rabbit IgG instead of HA723921 in mouse spleen tissue lysate

Blocking/Dilution buffer: primary antibody dilution (K1803)

Exposure time: 59 seconds; ECL: K1801

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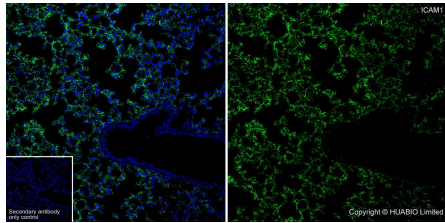


Fig7: This data was developed using HA723921, the same antibody clone in a different buffer formulation.

Application: Immunofluorescence (IF-tissue)

Species: Mouse

Tissue: Lung

Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST

Blocking: 10% normal goat serum + 1% Triton X-100 + 0.3 M Glycine in TBST, 30 minutes at room temperature.

Primary antibody: HA723921, 1/500, overnight at 4°C.

Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 1.5 hours at room temperature.

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

Background References

1. Grover JR et al. Basic motifs target PSGL-1, CD43, and CD44 to plasma membrane sites where HIV-1 assembles. J Virol 89:454-67 (2015).
2. Rouault C et al. Roles of chemokine ligand-2 (CXCL2) and neutrophils in influencing endothelial cell function and inflammation of human adipose tissue. Endocrinology 154:1069-79 (2013).

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