

# Anti-ICAM3 Antibody [PSH20-97] - BSA and Azide free

## HA751779



<b>Product Type:</b>	Recombinant Rabbit monoclonal IgG, primary antibodies
<b>Species reactivity:</b>	Human
<b>Applications:</b>	WB, IF-Cell, IHC-P, FC, IP
<b>Molecular Wt:</b>	Predicted band size: 60 kDa
<b>Clone number:</b>	PSH20-97

**Description:** Intercellular adhesion molecule 3 (ICAM3) also known as CD50 (Cluster of Differentiation 50), is a protein that in humans is encoded by the ICAM3 gene. The protein is constitutively expressed on the surface of leukocytes, which are also called white blood cells and are part of the immune system. ICAM3 mediates adhesion between cells by binding to specific integrin receptors. It plays an important role in the immune cell response through its facilitation of interactions between T cells and dendritic cells, which allows for T cell activation. ICAM3 also mediates the clearance of cells undergoing apoptosis by attracting and binding macrophages, a type of cell that breaks down infected or dying cells through a process known as phagocytosis, to apoptotic cells.

**Immunogen:** Recombinant protein within human ICAM3 aa 1-485.

**Positive control:** U-937 (Human acute monocytic leukemia cells) cell lysate, Jurkat (Human T-lymphoblastic cells) cell lysate, Ramos (Human Burkitt's lymphoma cells) cell lysate, THP-1 (Human acute monoblastic leukemia cells) cell lysate, HL-60 (Human acute promyelocytic leukemia cells) cell lysate.

**Subcellular location:** Membrane.

**Database links:** SwissProt: P32942 Human

**Recommended Dilutions:**

<b>WB</b>	1:5,000
<b>IF-Cell</b>	1:500
<b>IHC-P</b>	1:1,000
<b>FC</b>	1:1,000
<b>IP</b>	1-2µg/sample

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

**Storage Instruction:** Store at +4°C after thawing. Aliquot store at -20°C. Avoid repeated freeze / thaw cycles.

**Purity:** Protein A affinity purified.

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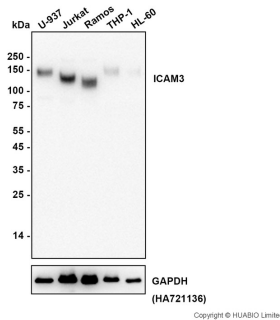
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## Images



**Fig1:** Western blot analysis of ICAM3 on different lysates with Rabbit anti-ICAM3 antibody (HA751779) at 1/5,000 dilution.

Lane 1: U-937 (Human acute monocytic leukemia cells) cell lysate  
 Lane 2: Jurkat (Human T-lymphoblastic cells) cell lysate  
 Lane 3: Ramos (Human Burkitt's lymphoma cells) cell lysate  
 Lane 4: THP-1 (Human acute monoblastic leukemia cells) cell lysate  
 Lane 5: HL-60 (Human acute promyelocytic leukemia cells) cell lysate

Lysates/proteins at 15 µg/Lane.

Exposure time: 20 seconds; ECL: K1801

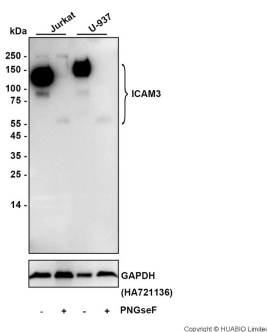
Blocking: 5% NFDm/TBST, 1 hour at room temperature

Primary antibody: HA751779, 1/5,000 in primary antibody dilution buffer (K1803), overnight at 4 °C

Secondary antibody: Goat anti-Rabbit IgG-HRP (HA1001), 1/50,000 in 5% NFDm/TBST, 1 hour at room temperature

Predicted band size: 60 kDa

Observed band size: 100-150 kDa



**Fig2:** Western blot analysis of ICAM3 on different lysates with Rabbit anti-ICAM3 antibody (HA751779) at 1/5,000 dilution.

Lane 1: Jurkat (Human T-lymphoblastic cells) cell lysate  
 Lane 2: Jurkat treated with PNGseF cell lysate  
 Lane 3: U-937 (Human acute monocytic leukemia cells) cell lysate  
 Lane 4: U-937 treated with PNGseF cell lysate

Lysates/proteins at 10 µg/Lane.

Exposure time: 3 minutes; ECL: K1801

Blocking: 5% NFDm/TBST, 1 hour at room temperature

Primary antibody: HA751779, 1/5,000 in primary antibody dilution buffer (K1803), overnight at 4 °C

Secondary antibody: Goat anti-Rabbit IgG-HRP (HA1001), 1/50,000 in 5% NFDm/TBST, 1 hour at room temperature

Predicted band size: 60 kDa

Observed band size: 100-150/60 kDa

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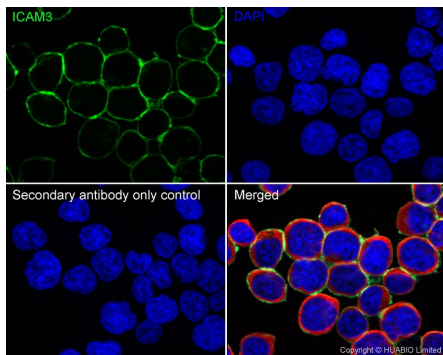
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**Fig3:** Application: Immunocytochemistry (IF-cell)

Species: Human

Sample: Jurkat (Human T-lymphoblastic cells)



Fixation: 4% Paraformaldehyde, 15 minutes at room temperature.  
 Permeabilization: 0.1% Triton X-100, 15 minutes at room temperature.

Blocking: 1% BSA + 10% normal goat serum, 1 hour at room temperature.

Antibody dilution buffer: 1% BSA in PBST.

Primary antibody: HA751779, 1/500, overnight at 4°C.

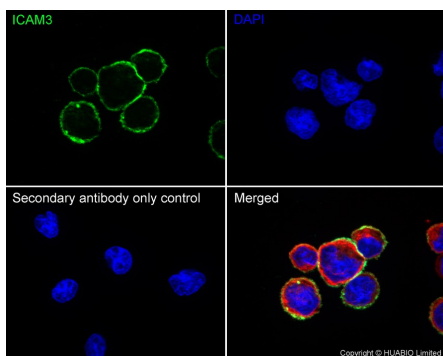
Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 45 minutes at room temperature.

Counterstain: Beta tubulin (HA601187, red), 1/100, overnight at 4°C. The Nuclear counterstain was DAPI (Blue).

**Fig4:** Application: Immunocytochemistry (IF-cell)

Species: Human

Sample: U-937 (Human acute monocytic leukemia cells)



Fixation: 4% Paraformaldehyde, 15 minutes at room temperature.  
 Permeabilization: 0.1% Triton X-100, 15 minutes at room temperature.

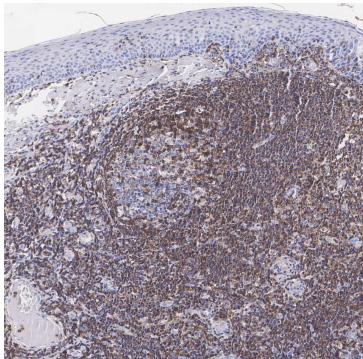
Blocking: 1% BSA + 10% normal goat serum, 1 hour at room temperature.

Antibody dilution buffer: 1% BSA in PBST.

Primary antibody: HA751779, 1/500, overnight at 4°C.

Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 45 minutes at room temperature.

Counterstain: Beta tubulin (HA601187, red), 1/100, overnight at 4°C. The Nuclear counterstain was DAPI (Blue).



**Fig5:** Application: Immunohistochemistry (IHC-P)

Species: Human

Tissue: Tonsil

Sample: Paraffin-embedded section

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

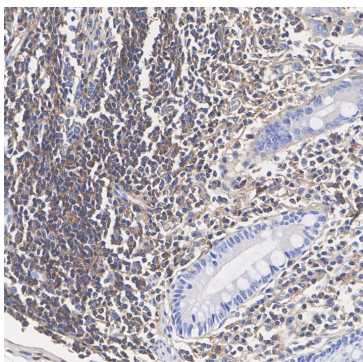
Wash buffer: 1× TBST

Endogenous peroxidase blocking: 3% H<sub>2</sub>O<sub>2</sub>, 10 minutes at room temperature.

Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.

Primary antibody: HA751779, 1/1,000, 1 hour at room temperature.

Secondary antibody: HA1119, 20 minutes at room temperature.



**Fig6:** Application: Immunohistochemistry (IHC-P)

Species: Human

Tissue: Colon

Sample: Paraffin-embedded section

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

Wash buffer: 1× TBST

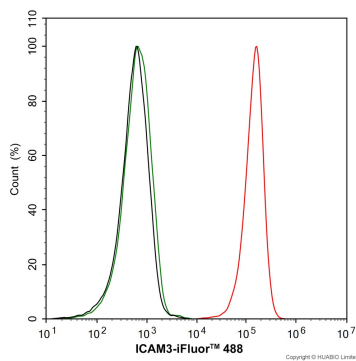
Endogenous peroxidase blocking: 3% H<sub>2</sub>O<sub>2</sub>, 10 minutes at room temperature.

Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.

Primary antibody: HA751779, 1/1,000, 1 hour at room temperature.

Secondary antibody: HA1119, 20 minutes at room temperature.

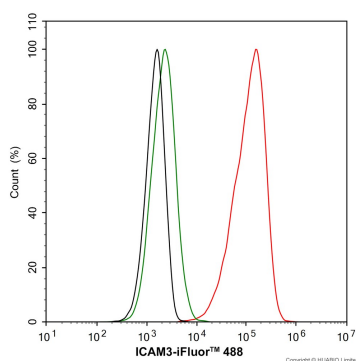
**Fig7:** Application: Flow Cytometry



Species: Human  
 Sample: Jurkat (Human T-lymphoblastic cells)

Blocking: 1% BSA + 10% normal goat serum + human FcR (Miltenyi Biotec), 15 minutes at room temperature. (human)  
 Antibody dilution buffer: 1x PBS.  
 Primary antibody: HA751779 (1/1,000) (Red) compared with Rabbit IgG Isotype Control (HA722127, Green), 15 minutes at room temperature.  
 Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 15 minutes at room temperature.

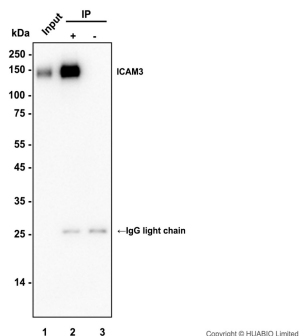
**Fig8:** Application: Flow Cytometry



Species: Human  
 Sample: U-937 (Human acute monocytic leukemia cells)

Blocking: 1% BSA + 10% normal goat serum + human FcR (Miltenyi Biotec), 15 minutes at room temperature. (human)  
 Antibody dilution buffer: 1x PBS.  
 Primary antibody: HA751779 (1/1,000) (Red) compared with Rabbit IgG Isotype Control (HA722127, Green), 15 minutes at room temperature.  
 Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 15 minutes at room temperature.

**Fig9:** Immunoprecipitation (IP)



ICAM3 was immunoprecipitated in 0.2 mg U-937 (Human acute monocytic leukemia cells) cell lysate with HA751779 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using HA751779 at 1/5,000 dilution. Anti-Rabbit IgG for IP Nano-secondary antibody (NBI01H) at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: U-937 cell lysate (input)  
 Lane 2: HA751779 IP in U-937 cell lysate  
 Lane 3: Rabbit IgG instead of HA751779 in U-937 cell lysate

Exposure time: 14 seconds  
 Blocking: 5% NFDM/TBST, 1 hour at room temperature

Primary dilution: HA751779, 1/5,000 in primary antibody dilution buffer (K1803), 2 hours at room temperature

Predicted band size: 60 kDa  
 Observed band size: 100-150 kDa

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

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### Background References

1. Li L et al. Functional evaluation of various ICAM3 transcript variants in diffuse large B-Cell lymphoma. *Leuk Lymphoma*. 2022 Dec
2. Zicheng H et al. Association of Circulating ICAM3 Concentrations with Severity and Short-term Outcomes of Acute Ischemic Stroke. *Neurotox Res*. 2021 Aug

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