Anti-CD166 Antibody [A6A9]

HA600082

Product Type:	Mouse monoclonal IgG2a, primary antibodies
Species reactivity:	Human
Applications:	IF-Cell, IHC-P, WB
Molecular Wt:	Predicted band size: 65 kDa.
Clone number:	A6A9

Description: This gene encodes activated leukocyte cell adhesion molecule (ALCAM), also known as CD166 (cluster of differentiation 166), which is a member of a subfamily of immunoglobulin receptors with five immunoglobulin-like domains (VVC2C2C2) in the extracellular domain. This protein binds to T-cell differentiation antigene CD6, and is implicated in the processes of cell adhesion and migration. Multiple alternatively spliced transcript variants encoding different isoforms have been found.

Immunogen: Recombinant protein within human CD166 aa 51-250.

Positive control: U-2 OS cell lysate, A549 cell lysate, LNCaP cell lysate, HepG2 cell lysate, A549, human endometrium tissue, human liver tissue, human stomach tissue.

Subcellular location: Cell membrane, axon, dendrite; Secreted.

Database links: SwissProt: Q13740 Human

Recommended Dilutions:

IF-Cell	1:100
IHC-P	1:1,000-1:2,000
WB	1:1,000

Storage Buffer: 1*TBS (pH7.4), 0.2% BSA, 50% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein G affinity purified.

Images

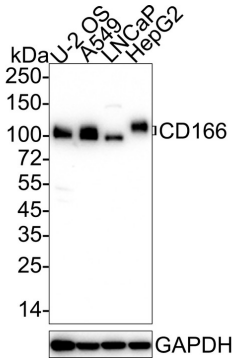

Fig1: Western blot analysis of CD166 on different lysates with Mouse anti-CD166 antibody (HA600082) at 1/1,000 dilution.

Lane 1: U-2 OS cell lysate
Lane 2: A549 cell lysate
Lane 3: LNCaP cell lysate
Lane 4: HepG2 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 65 kDa
Observed band size: 100-105 kDa

Exposure time: 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 (HA600082) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

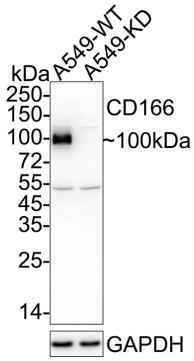

Fig2: Western blot analysis of CD166 on different lysates with Mouse anti-CD166 antibody (HA600082) at 1/2,000 dilution.

Lane 1: A549-WT cell lysate
Lane 2: A549-KD CD166 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 65 kDa
Observed band size: 100 kDa

Exposure time: 1 minutes 36 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 (HA600082) at 1/2,000 dilution was used in 5% BSA at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

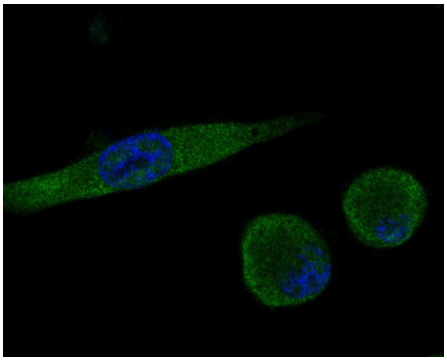

Fig3: ICC staining of CD166 in A549 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (HA600082, 1/100) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 conjugate-Goat anti-Mouse IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

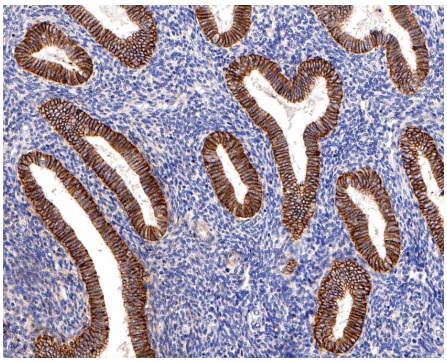

Fig4: Immunohistochemical analysis of paraffin-embedded human endometrium tissue using anti-CD166 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA600082, 1/800) for 30 minutes 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.

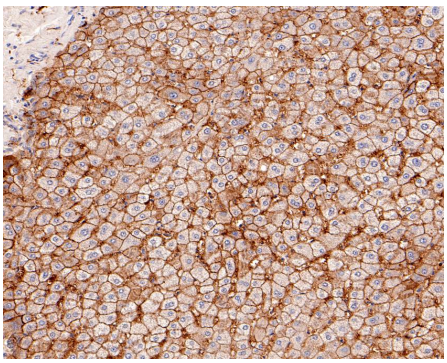

Fig5: Immunohistochemical analysis of paraffin-embedded human liver tissue using anti-CD166 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA600082, 1/1,500) for 30 minutes 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.

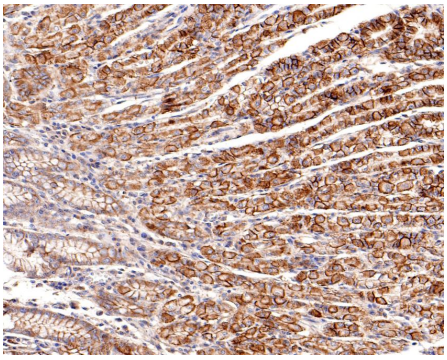

Fig6: Immunohistochemical analysis of paraffin-embedded human stomach tissue using anti-CD166 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA600082, 1/800) for 30 minutes 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.

Background References

1. Münsterberg J. et. al. ALCAM contributes to brain metastasis formation in non-small-cell lung cancer through interaction with the vascular endothelium. Neuro Oncol. 2020 Jul
2. Bartolomé RA. et. al. SOSTDC1 promotes invasion and liver metastasis in colorectal cancer via interaction with ALCAM/CD166. Oncogene. 2020 Sep