Anti-ALDH7A1 Antibody [JE63-38]

HA720086

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IF-Cell, IHC-P, FC
Molecular Wt:	Predicted band size: 58 kDa
Clone number:	JE63-38

Description: The protein encoded by this gene is a member of subfamily 7 in the aldehyde dehydrogenase gene family. These enzymes are thought to play a major role in the detoxification of aldehydes generated by alcohol metabolism and lipid peroxidation. This particular member has homology to a previously described protein from the green garden pea, the 26g pea turgor protein. It is also involved in lysine catabolism that is known to occur in the mitochondrial matrix. Recent reports show that this protein is found both in the cytosol and the mitochondria, and the two forms likely arise from the use of alternative translation initiation sites. An additional variant encoding a different isoform has also been found for this gene. Mutations in this gene are associated with pyridoxine-dependent epilepsy. Several related pseudogenes have also been identified.

Immunogen: Synthetic peptide within human ALDH7A1 aa 36-85/539.

Positive control: Hela cell lysate, HepG2 cell lysate, 293T cell lysate, A431 cell lysate, Mouse liver tissue lysate, Mouse kidney tissue lysate, A549 cell lysate, Hela, human liver tissue, human kidney tissue, mouse brain tissue, mouse kidney tissue, HepG2.

Subcellular location: Cytoplasm, Mitochondrion, Nucleus.

Database links: SwissProt: P49419 Human | Q9DBF1 Mouse

Recommended Dilutions:

WB	1:500
IF-Cell	1:50
IHC-P	1:500-1:1,000
FC	1:500-1:1,000

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% 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 A affinity purified.

Images

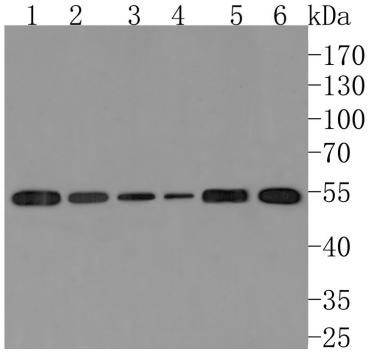

Fig1: Western blot analysis of ALDH7A1 on different lysates with Rabbit anti-ALDH7A1 antibody (HA720086) at 1/500 dilution.

Lane 1: Hela cell lysate, 10 µg/Lane
Lane 2: HepG2 cell lysate, 10 µg/Lane
Lane 3: 293T cell lysate, 10 µg/Lane
Lane 4: A431 cell lysate, 10 µg/Lane
Lane 5: Mouse liver tissue lysate, 20 µg/Lane
Lane 6: Mouse kidney tissue lysate, 20 µg/Lane

Predicted band size: 58 kDa
Observed band size: 55 kDa

Exposure time: 2 minutes;

10% 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 (HA720086) at 1/500 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

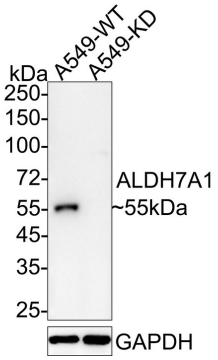

Fig2: Western blot analysis of ALDH7A1 on different lysates with Rabbit anti-ALDH7A1 antibody (HA720086) at 1/2,000 dilution.

Lane 1: A549-si NT cell lysate
Lane 2: A549-si ALDH7A1 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 58 kDa
Observed band size: 55 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 (HA720086) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

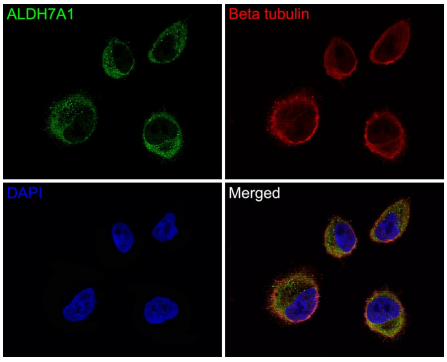

Fig3: Immunocytochemistry analysis of Hela cells labeling ALDH7A1 with Rabbit anti-ALDH7A1 antibody (HA720086) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 ℃, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-ALDH7A1 antibody (HA720086) at 1/50 dilution in 2% negative goat serum overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) was used as the secondary antibody at 1/1,000 dilution. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/200 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (Alexa Fluor® 647) were used as the secondary antibody at 1/1,000 dilution.

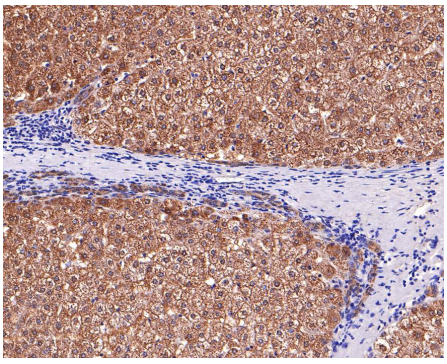

Fig4: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-ALDH7A1 antibody (HA720086) at 1/1,000 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (HA720086) at 1/1,000 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.

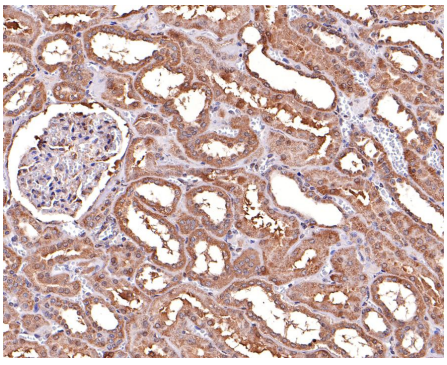

Fig5: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-ALDH7A1 antibody (HA720086) at 1/500 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (HA720086) at 1/500 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.

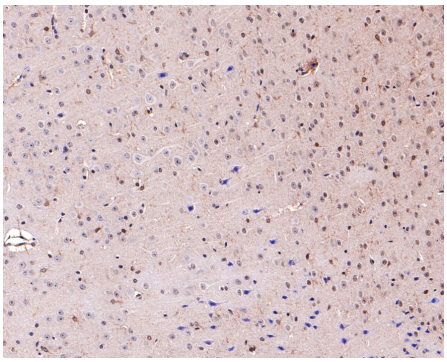

Fig6: Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Rabbit anti-ALDH7A1 antibody (HA720086) at 1/1,000 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (HA720086) at 1/1,000 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.

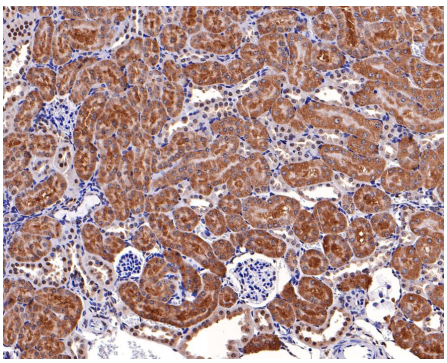

Fig7: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-ALDH7A1 antibody (HA720086) at 1/500 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (HA720086) at 1/500 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.

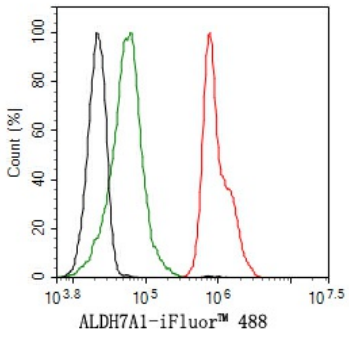

Fig8: Flow cytometric analysis of HepG2 cells labeling ALDH7A1.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA720086, 1ug/ml) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ 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℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Coughlin CR. et. al. The genotypic spectrum of ALDH7A1 mutations resulting in pyridoxine dependent epilepsy: A common epileptic encephalopathy. J Inherit Metab Dis. 2019 Mar
2. Toldo I. et. al. Brain malformations associated to Aldh7a1 gene mutations: Report of a novel homozygous mutation and literature review. Eur J Paediatr Neurol. 2018 Nov