Anti-HADHSC Antibody [D10-E7]

M1409-2

Product Type:	Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 34 kDa
Clone number:	D10-E7

Description: Hydroxyacyl-Coenzyme A dehydrogenase also known as HADH is an enzyme which in humans is encoded by the HADH gene. This gene is a member of the 3-hydroxyacyl-CoA dehydrogenase gene family. The encoded protein functions in the mitochondrial matrix to catalyze the oxidation of straight-chain 3-hydroxyacyl-CoAs as part of the beta-oxidation pathway. Its enzymatic activity is highest with medium-chain-length fatty acids. Mutations in this gene cause one form of familial hyperinsulinemic hypoglycemia. A deficiency is associated with 3-hydroxyacyl-coenzyme A dehydrogenase deficiency.

Immunogen: Synthetic peptide within Human HADHSC aa 265-314 / 314.

Positive control: HepG2 cell lysate, HeLa cell lysate, HT-29 cell lysate, HL-60 cell lysate, A431 cell lysate, K-562 cell lysate, human liver tissue lysate, mouse liver tissue lysate, mouse heart tissue lysate, rat liver tissue lysate, rat heart tissue lysate, zebrafish tissue lysate, HeLa, human kidney tissue, human colon carcinoma tissue, human liver tissue, mouse liver tissue, rat liver tissue.

Subcellular location: Mitochondrion matrix.

Database links: SwissProt: Q16836 Human | Q61425 Mouse | Q9WVK7 Rat

Recommended Dilutions:

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

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

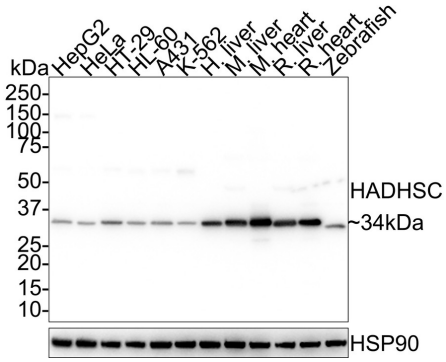

Fig1: Western blot analysis of HADHSC on different lysates with Mouse anti-HADHSC antibody (M1409-2) at 1/1,000 dilution.

Lane 1: HepG2 cell lysate (20 µg/Lane)
Lane 2: HeLa cell lysate (20 µg/Lane)
Lane 3: HT-29 cell lysate (20 µg/Lane)
Lane 4: HL-60 cell lysate (20 µg/Lane)
Lane 5: A431 cell lysate (20 µg/Lane)
Lane 6: K-562 cell lysate (20 µg/Lane)
Lane 7: Human liver tissue lysate (40 µg/Lane)
Lane 8: Mouse liver tissue lysate (40 µg/Lane)
Lane 9: Mouse heart tissue lysate (40 µg/Lane)
Lane 10: Rat liver tissue lysate (40 µg/Lane)
Lane 11: Rat heart tissue lysate (40 µg/Lane)
Lane 12: Zebrafish tissue lysate (40 µg/Lane)

Predicted band size: 34 kDa
Observed band size: 34 kDa

Exposure time: 5 seconds;

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 (M1409-2) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

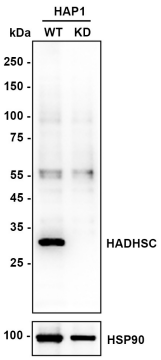

Fig2: Western blot analysis of HADHSC on different lysates with Mouse anti-HADHSC antibody (M1409-2) at 1/2,000 dilution.

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

Lysates/proteins at 10 µg/Lane.

Predicted band size: 34 kDa
Observed band size: 34 kDa

Exposure time: 9 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 (M1409-2) at 1/2,000 dilution was used in K1803 at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody

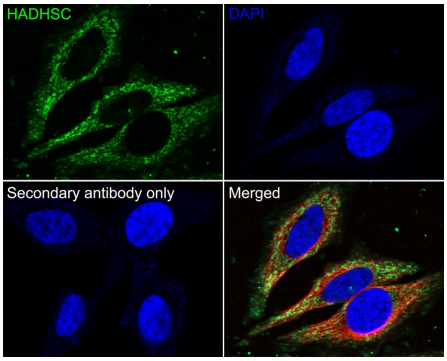

Fig3: Immunocytochemistry analysis of HeLa cells labeling HADHSC with Mouse anti-HADHSC antibody (M1409-2) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 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-HADHSC antibody (M1409-2) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 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℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

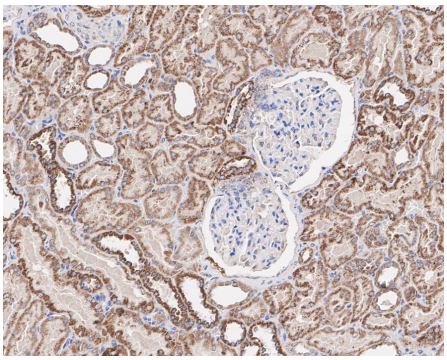

Fig4: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Mouse anti-HADHSC antibody (M1409-2) at 1/1,000 dilution.

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 (M1409-2) 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.

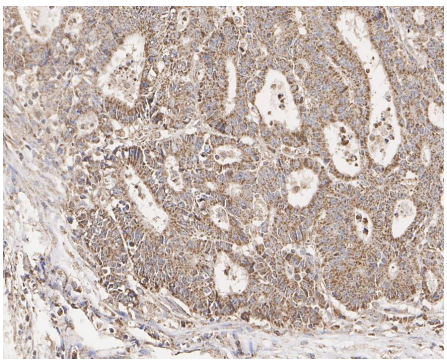

Fig5: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue with Mouse anti-HADHSC antibody (M1409-2) at 1/1,000 dilution.

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 (M1409-2) 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.

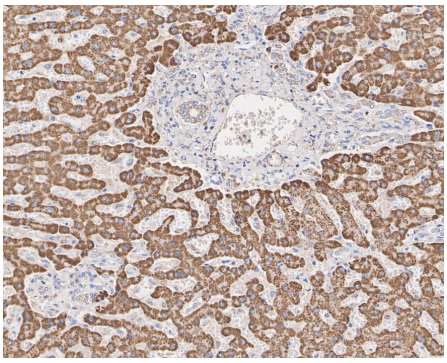

Fig6: Immunohistochemical analysis of paraffin-embedded human liver tissue with Mouse anti-HADHSC antibody (M1409-2) at 1/1,000 dilution.

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 (M1409-2) 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.

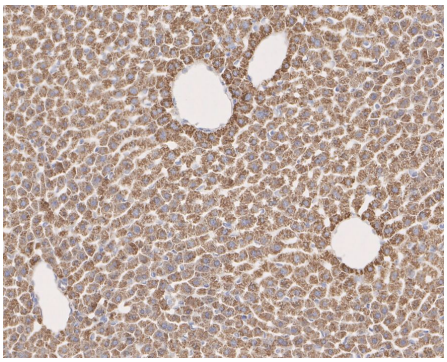

Fig7: Immunohistochemical analysis of paraffin-embedded mouse liver tissue with Mouse anti-HADHSC antibody (M1409-2) at 1/1,000 dilution.

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 (M1409-2) 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.

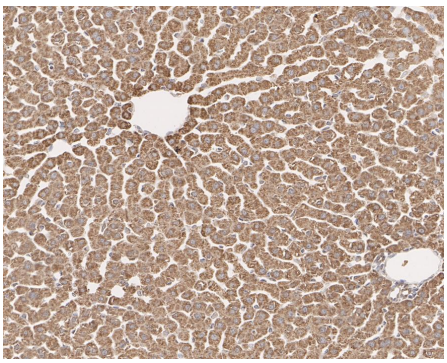

Fig8: Immunohistochemical analysis of paraffin-embedded rat liver tissue with Mouse anti-HADHSC antibody (M1409-2) at 1/1,000 dilution.

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 (M1409-2) 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.

Background References

1. "Sequestration of the active site by interdomain shifting. Crystallographic and spectroscopic evidence for distinct conformations of L-3-hydroxyacyl-CoA dehydrogenase." Barycki J.J., O'Brien L.K., Strauss A.W., Banaszak L.J. J. Biol. Chem. 275:27186-27196(2000)
2. "Fulminant hepatic failure associated with mutations in the medium and short chain L-3-hydroxyacyl-CoA dehydrogenase gene." O'Brien L.K., Rinaldo P., Sims H.F., Alonso E.M., Charrow J., Jones P.M., Bennett M.J., Barycki J.J., Banaszak L.J., Strauss A.W. J. Inherit. Metab. Dis. 23 Suppl. 1:127-127(2000)
3. "3-hydroxyacyl-CoA dehydrogenase and short chain 3-hydroxyacyl-CoA dehydrogenase in human health and

disease." Yang S.-Y., He X.-Y., Schulz H. FEBS J. 272:4874-4883(2005)