Anti-HCLS1 Antibody [PSH01-12]

HA721624

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IHC-P, IF-Cell, FC
Molecular Wt:	Predicted band size: 54 kDa
Clone number:	PSH01-12

Description: Hematopoietic lineage cell-specific protein is a protein that in humans is encoded by the HCLS1 gene. Substrate of the antigen receptor-coupled tyrosine kinase. Plays a role in antigen receptor signaling for both clonal expansion and deletion in lymphoid cells. May also be involved in the regulation of gene expression. Enables RNA polymerase II-specific DNA-binding transcription factor binding activity and protein kinase binding activity. Involved in several processes, including positive regulation of intracellular signal transduction; positive regulation of protein phosphorylation; and regulation of transcription, DNA-templated. Located in cytosol; nucleus; and plasma membrane. Part of transcription regulator complex. HCLS1 has been shown to interact with Caspase 3.

Immunogen: Recombinant protein within human HCLS1 aa 201-450 / 486

Positive control: Ramos cell lysate, Daudi cell lysate, Raji cell lysate, Jurkat cell lysate, HL-60 cell lysate, human lung carcinoma tissue, human liver tissue, human brain tissue, HL-60, Jurkat.

Subcellular location: Membrane, Cytoplasm, Mitochondrion.

Database links: SwissProt: P14317 Human

Recommended Dilutions:

WB	1:2,000
IHC-P	1:200-1:5,000
IF-Cell	1:100-1:250
FC	1:500-1:1,000

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

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

Purity: Protein A affinity purified.

Images

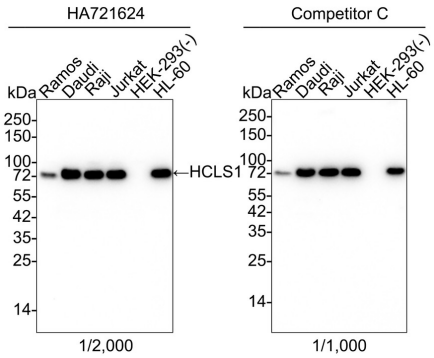

Fig1: Western blot analysis of HCLS1 on different lysates with Rabbit anti-HCLS1 antibody (HA721624) at 1/2,000 dilution and competitor's antibody at 1/1,000 dilution.

Lane 1: Ramos cell lysate (20 µg/Lane)
Lane 2: Daudi cell lysate (20 µg/Lane)
Lane 3: Raji cell lysate (20 µg/Lane)
Lane 4: Jurkat cell lysate (20 µg/Lane)
Lane 5: HEK-293 cell lysate (negative) (20 µg/Lane)
Lane 6: HL-60 cell lysate (20 µg/Lane)

Predicted band size: 54 kDa
Observed band size: 75 kDa

Exposure time: 42 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 (HA721624) at 1/2,000 dilution and competitor's antibody at 1/1,000 dilution were 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.

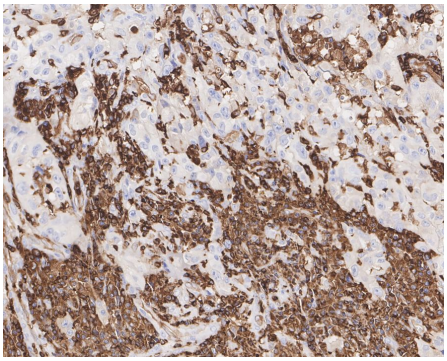

Fig2: Immunohistochemical analysis of paraffin-embedded human lung carcinoma tissue with Rabbit anti-HCLS1 antibody (HA721624) at 1/5,000 dilution.

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 (HA721624) at 1/5,000 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.

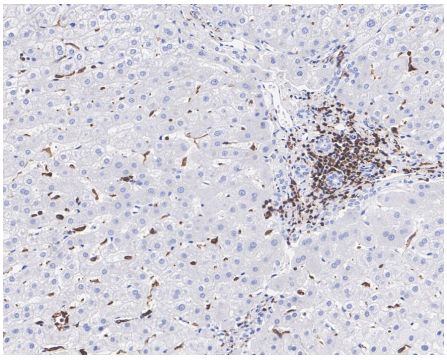

Fig3: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-HCLS1 antibody (HA721624) at 1/1,000 dilution.

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 (HA721624) 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.

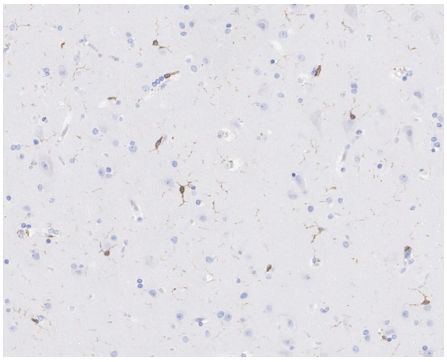

Fig4: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-HCLS1 antibody (HA721624) at 1/200 dilution.

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 (HA721624) at 1/200 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.

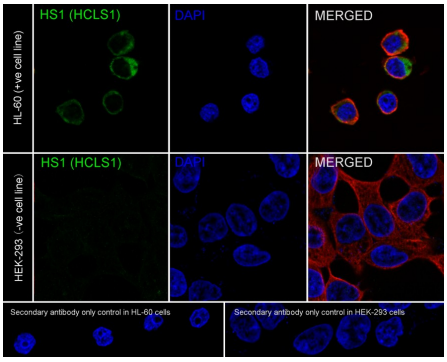

Fig5: Immunocytochemistry analysis of HL-60 (positive) and HEK-293 (negative) labeling HCLS1 with Rabbit anti-HCLS1 antibody (HA721624) at 1/100 dilution.

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 Rabbit anti-HCLS1 antibody (HA721624) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. 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 (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

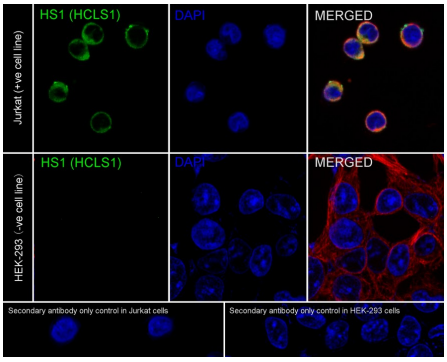

Fig6: Immunocytochemistry analysis of Jurkat (positive) and HEK-293 (negative) labeling HCLS1 with Rabbit anti-HCLS1 antibody (HA721624) at 1/100 dilution.

Cells were fixed in 100% precooled methanol 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 Rabbit anti-HCLS1 antibody (HA721624) at 1/250 dilution in 1% BSA in PBST overnight at 4 ℃. 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 (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Background References

1. Quatrana A et al. Hsa-miR223-3p circulating level is upregulated in Friedreich's ataxia and inversely associated with HCLS1 associated protein X-1, HAX-1. Hum Mol Genet. 2022 Jun