Anti-EIF2S1 Antibody [PSH04-29]

HA722112

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat, Monkey
Applications:	WB, IF-Cell, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 36 kDa
Clone number:	PSH04-29

Description: Eukaryotic translation initiation factor 2 subunit 1 (eIF2α) is a protein that in humans is encoded by the EIF2S1 gene. The protein encoded by this gene is the alpha (α) subunit of the translation initiation factor eIF2 protein complex which catalyzes an early regulated step of protein synthesis initiation, promoting the binding of the initiator tRNA (Met-tRNAiMet) to 40S ribosomal subunits. Binding occurs as a ternary complex of methionyl-tRNA, eIF2, and GTP. eIF2 is composed of 3 nonidentical subunits, alpha (α, 36 kD, this article), beta (β, 38 kD), and gamma (γ, 52 kD). The rate of formation of the ternary complex is modulated by the phosphorylation state of eIF2α. Phosphorylation of eIF2α by EIF-2 kinases plays a key role in regulating the integrated stress response.

Immunogen: Recombinant protein within human EIF2S1 aa 1-315 / 315.

Positive control: MCF7 cell lysate, HepG2 cell lysate, HeLa cell lysate, COS-1 cell lysate, A549 cell lysate, RAW264.7 cell lysate, C6 cell lysate, mouse kidney tissue lysate, mouse spleen tissue lysate, rat kidney tissue lysate, rat spleen tissue lysate, human breast tissue, mouse breast tissue, rat breast tissue, HepG2.

Subcellular location: Cytoplasm, Stress granule, cytosol, Mitochondrion.

Database links: SwissProt: P05198 Human | Q6ZWX6 Mouse | P68101 Rat

Recommended Dilutions:

WB	1:2,000
IF-Cell	1:100
IHC-P	1:1,000
FC	1:1,000
IP	1-2μg/sample

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

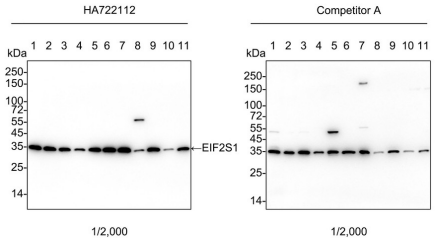

Fig1: Western blot analysis of EIF2S1 on different lysates with Rabbit anti-EIF2S1 antibody (HA722112) at 1/2,000 dilution and competitor's antibody at 1/2,000 dilution.

Lane 1: MCF7 cell lysate
Lane 2: HepG2 cell lysate
Lane 3: HeLa cell lysate
Lane 4: COS-1 cell lysate
Lane 5: A549 cell lysate
Lane 6: RAW264.7 cell lysate
Lane 7: C6 cell lysate
Lane 8: Mouse kidney tissue lysate
Lane 9: Mouse spleen tissue lysate
Lane 10: Rat kidney tissue lysate
Lane 11: Rat spleen tissue lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 36 kDa
Observed band size: 36 kDa

Exposure time: 59 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 (HA722112) at 1/2,000 dilution and competitor's antibody at 1/2,000 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.

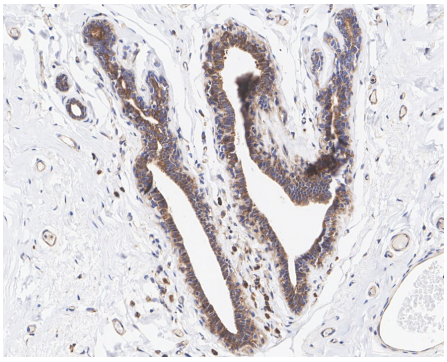

Fig2: Immunohistochemical analysis of paraffin-embedded human breast tissue with Rabbit anti-EIF2S1 antibody (HA722112) at 1/1,000 dilution.

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 (HA722112) 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.

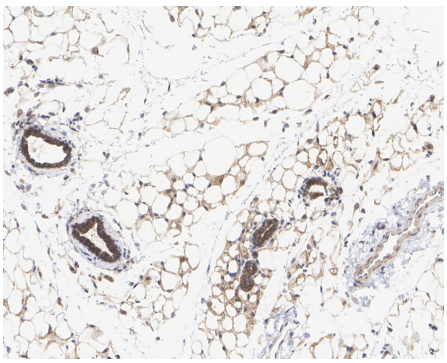

Fig3: Immunohistochemical analysis of paraffin-embedded mouse breast tissue with Rabbit anti-EIF2S1 antibody (HA722112) at 1/1,000 dilution.

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 (HA722112) 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.

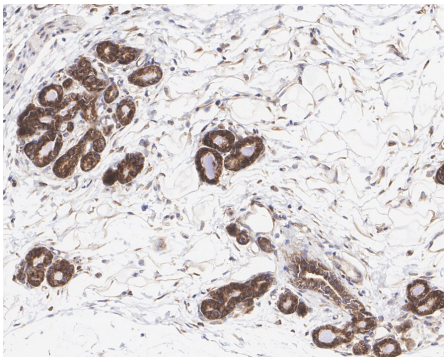

Fig4: Immunohistochemical analysis of paraffin-embedded rat breast tissue with Rabbit anti-EIF2S1 antibody (HA722112) at 1/1,000 dilution.

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 (HA722112) 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.

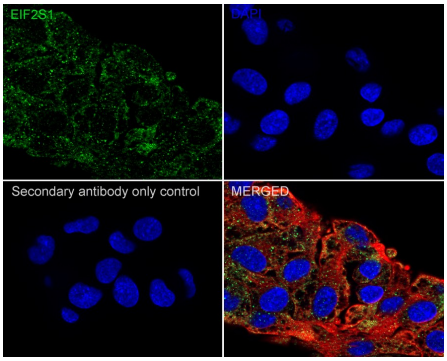

Fig5: Immunocytochemistry analysis of HepG2 cells labeling EIF2S1 with Rabbit anti-EIF2S1 antibody (HA722112) at 1/100 dilution.

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-EIF2S1 antibody (HA722112) 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.

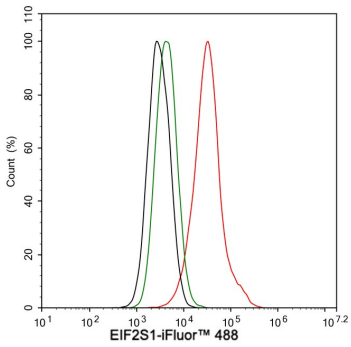

Fig6: Flow cytometric analysis of HepG2 cells labeling EIF2S1.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA722112, 1μg/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).

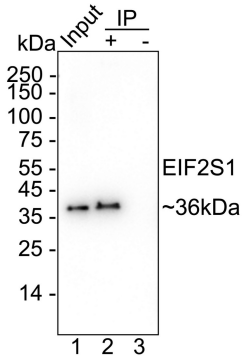

Fig7: EIF2S1 was immunoprecipitated from 0.2 mg HepG2 cell lysate with HA722112 at 2 μg/10 μl beads. Western blot was performed from the immunoprecipitate using HA722112 at 1/2,000 dilution. HRP Conjugated Anti-Rabbit IgG for IP Nano-secondary antibody at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: HepG2 cell lysate (input)
Lane 2: HA722112 IP in HepG2 cell lysate
Lane 3: Rabbit IgG instead of HA722112 in HepG2 cell lysate

Blocking/Dilution buffer: primary antibody dilution (K1803)
Exposure time: 2 seconds; ECL: K1801

Background References

1. Dang TT et al. Phosphorylation of EIF2S1 (eukaryotic translation initiation factor 2 subunit alpha) is indispensable for nuclear translocation of TFEB and TFE3 during ER stress. Autophagy. 2023 Jul
2. Shi JX et al. MiR-3074-5p Regulates Trophoblasts Function via EIF2S1/GDF15 Pathway in Recurrent Miscarriage. Reprod Sci. 2023 Dec