Anti-STUB1 Antibody [JG38-22]

ET7108-65



Product Type: Recombinant Rabbit monoclonal IgG, primary antibodies

Species reactivity: Human, Mouse, Rat

Applications: WB, IF-Cell, IHC-P, IP, FC

Molecular Wt: Predicted band size: 35 kDa

Clone number: JG38-22

Description: CHIP (carboxy terminus of HSP 70-interacting protein), also designated STIP1 homology

and U-box containing protein 1, HSPABP2, NY-CO-7, SDCCAG7 and STUB1, is a cytoplasmic E3 ubiquitin ligase that influences protein ubiquitylation. CHIP interacts with Smad1/Smad4 and blocks BMP signaling through the ubiquitin-mediated degradation of Smad proteins. CHIP controls both association of Hsp70/Hsp90 chaperones with ErbB2 and down-regulation of ErbB2 induced by inhibitors of Hsp90. A 1.3-kb transcript is most abundant in striated muscle (heart and skeletal muscle), with lower expression in pancreas

and brain.

Immunogen: Synthetic peptide within Human STUB1 aa 10-59 / 303.

Positive control: HeLa cell lysate, HEK-293 cell lysate, human brain tissue lysate, human heart tissue, mouse

heart tissue, rat heart tissue, 293T, LOVO, MCF-7, SH-SY5Y.

Subcellular location: Nucleus. Cytoplasm.

Database links: SwissProt: Q9UNE7 Human | Q9WUD1 Mouse

Entrez Gene: 287155 Rat

Recommended Dilutions:

WB 1:1,000-1:2,000
IF-Cell 1:50-1:200
IHC-P 1:50-1:200
FC 1:50-1:100

IP Use at an assay dependent concentration.

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°C long term.

Purity: Protein A affinity purified.

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Images

kDa ke kiki k 250-150-100-72-55-45-35-25-14-HSP90 **Fig1:** Western blot analysis of STUB1 on different lysates with Rabbit anti-STUB1 antibody (ET7108-65) at 1/1,000 dilution.

Lane 1: HeLa cell lysate (15 µg/Lane) Lane 2: HEK-293 cell lysate (15 µg/Lane) Lane 3: Human brain tissue lysate (30 µg/Lane)

Predicted band size: 35 kDa Observed band size: 34 kDa

Exposure time: 14 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

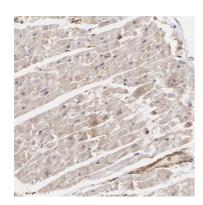


Fig2: Immunohistochemical analysis of paraffin-embedded human heart tissue with Rabbit anti-STUB1 antibody (ET7108-65) at 1/200 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (ET7108-65) 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.

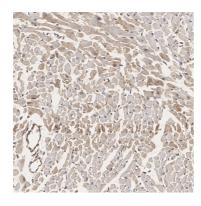


Fig3: Immunohistochemical analysis of paraffin-embedded mouse heart tissue with Rabbit anti-STUB1 antibody (ET7108-65) at 1/200 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (ET7108-65) 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.

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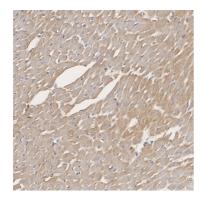


Fig4: Immunohistochemical analysis of paraffin-embedded rat heart tissue with Rabbit anti-STUB1 antibody (ET7108-65) at 1/200 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (ET7108-65) 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.

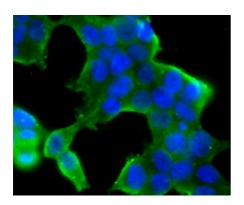


Fig5: ICC staining of STUB1 in 293T cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET7108-65, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

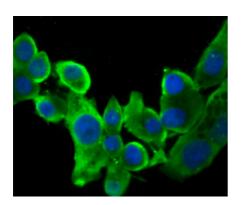


Fig6: ICC staining of STUB1 in LOVO cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET7108-65, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

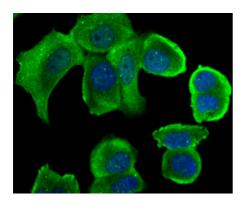


Fig7: ICC staining of STUB1 in MCF-7 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET7108-65, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

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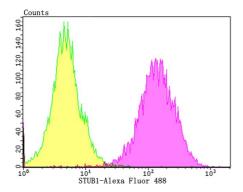


Fig8: Flow cytometric analysis of STUB1 was done on SH-SY5Y cells. The cells were fixed, permeabilized and stained with the primary antibody (ET7108-65, 1/50) (purple). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes.Unlabelled sample was used as a control (cells without incubation with primary antibody; yellow).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

- 1. Ballinger C A et al. Identification of CHIP, a novel tetratricopeptide repeat-containing protein that interacts with heat shock proteins and negatively regulates chaperone functions. Mol Cell Biol 19:4535-4545 (1999).
- 2. Jiang J et al. CHIP is a U-box-dependent E3 ubiquitin ligase: identification of Hsc70 as a target for ubiquitylation. J Biol Chem 276:42938-42944 (2001).