Anti-ATG7 Antibody [SC06-30]

ET1610-53

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Rat
Applications:	WB, IF-Tissue, IHC-P, IP, FC
Molecular Wt:	Predicted band size: 78 kDa
Clone number:	SC06-30

Description: E1-like activating enzyme involved in the 2 ubiquitin-like systems required for cytoplasm to vacuole transport (Cvt) and autophagy. Activates ATG12 for its conjugation with ATG5 as well as the ATG8 family proteins for their conjugation with phosphatidylethanolamine. Both systems are needed for the ATG8 association to Cvt vesicles and autophagosomes membranes. Required for autophagic death induced by caspase-8 inhibition. Required for mitophagy which contributes to regulate mitochondrial quantity and quality by eliminating the mitochondria to a basal level to fulfill cellular energy requirements and preventing excess ROS production. Modulates p53/TP53 activity to regulate cell cycle and survival during metabolic stress. Plays also a key role in the maintenance of axonal homeostasis, the prevention of axonal degeneration, the maintenance of hematopoietic stem cells, the formation of Paneth cell granules, as well as in adipose differentiation. Plays a role in regulating the liver clock and glucose metabolism by mediating the autophagic degradation of CRY1 (clock repressor) in a time-dependent manner.

Immunogen: Synthetic peptide within Human ATG7 aa 654-703 / 703.

Positive control: HeLa cell lysate, Jurkat cell lysate, THP-1 cell lysate, human tonsil tissue, human spleen tissue, human kidney tissue, human liver tissue, rat kidney tissue lysate.

Subcellular location: Cytoplasm.

Database links: SwissProt: O95352 Human | Q641Y5 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IF-Tissue	1:50-1:200
IHC-P	1:50-1:1,000
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: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

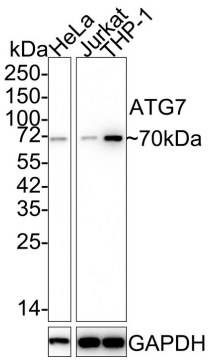

Fig1: Western blot analysis of ATG7 on different lysates with Rabbit anti-ATG7 antibody (ET1610-53) at 1/2,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: Jurkat cell lysate
Lane 3: THP-1 cell lysate

Lysates/proteins at 15 µg/Lane.

Predicted band size: 78 kDa
Observed band size: 70 kDa

Exposure time: 3 minutes;

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 (ET1610-53) 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.

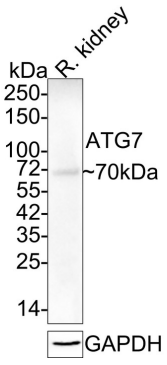

Fig2: Western blot analysis of ATG7 on rat kidney tissue lysates with Rabbit anti-ATG7 antibody (ET1610-53) at 1/1,000 dilution.

Lysates/proteins at 40 µg/Lane.

Predicted band size: 78 kDa
Observed band size: 70 kDa

Exposure time: 3 minutes;
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 (ET1610-53) at 1/1,000 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.

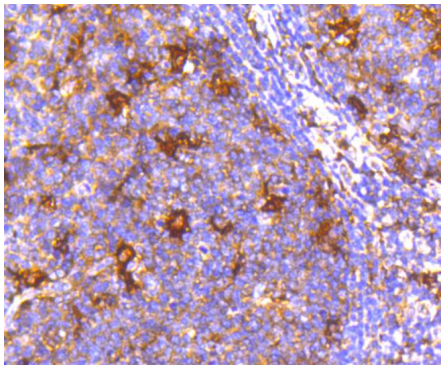

Fig3: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-Atg7 antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1610-53, 1/50) for 30 minutes 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.

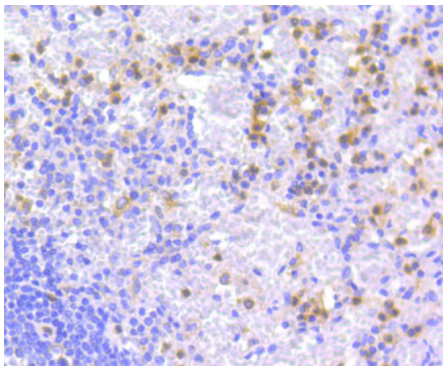

Fig4: Immunohistochemical analysis of paraffin-embedded human spleen tissue using anti-Atg7 antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1610-53, 1/50) for 30 minutes 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.

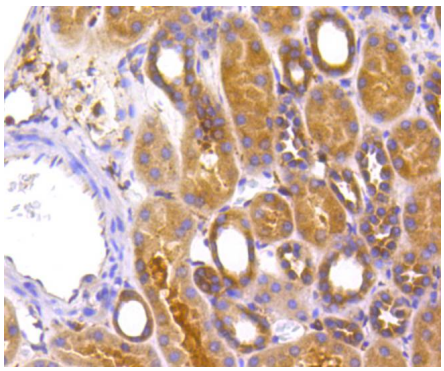

Fig5: Immunohistochemical analysis of paraffin-embedded human kidney tissue using anti-Atg7 antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1610-53, 1/50) for 30 minutes 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.

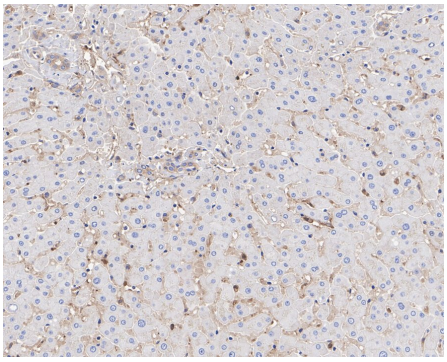

Fig6: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-ATG7 antibody (ET1610-53) at 1/1,000 dilution.

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 (ET1610-53) 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.

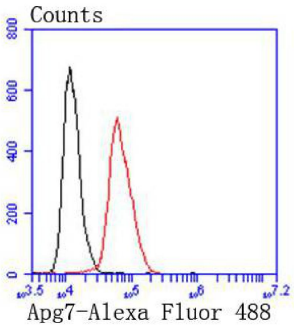

Fig7: Flow cytometric analysis of Atg7 was done on Hela cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1610-53, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes.Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

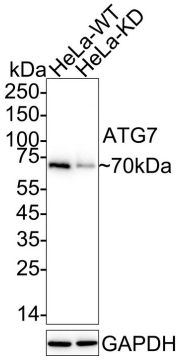

Fig8: Western blot analysis of ATG7 on different lysates with Rabbit anti-ATG7 antibody (ET1610-53) at 1/2,000 dilution.

Lane 1: HeLa-si NT cell lysate
Lane 2: HeLa-si ATG7 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 78 kDa
Observed band size: 70 kDa

Exposure time: 10 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 (ET1610-53) 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.

Background References

1. He G et al. Gadd45b prevents autophagy and apoptosis against rat cerebral neuron oxygen-glucose deprivation/reperfusion injury. Apoptosis 21:390-403 (2016).
2. Nazarko TY et al. Peroxisomal Atg37 binds Atg30 or palmitoyl-CoA to regulate phagophore formation during pexophagy. J Cell Biol 204:541-57 (2014).