Anti-MAP1LC3A Antibody [ST47-03]

ET1609-26

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 14 kDa
Clone number:	ST47-03

Description: Microtubules, the primary component of the cytoskeletal network, interact with proteins called microtubule-associated proteins (MAPs). The microtubule-associated proteins can be divided into two groups, structural and dynamic. The structural microtubule-associated proteins, MAP-1A, MAP-1B, MAP-2A, MAP-2B and MAP-2C, stimulate tubulin assembly, enhance micro-tubule stability and influence the spatial distribution of microtubules within cells. Both MAP-1 and, to a greater extent, MAP-2 have been implicated as agents of microtubule depolymerization by suppressing the dynamic instability of the microtubules. The suppression of microtubule dynamic instability by the MAP proteins is thought to be associated with phosphorylation of the MAPs.

Immunogen: Synthetic peptide within Human MAP1LC3A aa 81-121 / 121.

Positive control: HeLa treated with 50μM Chloroquine for 18 hours cell lysate, C2C12 cell lysate, C2C12 treated with 50μM Chloroquine for 18 hours cell lysate, C6 cell lysate, C6 treated with 50μM Chloroquine for 18 hours cell lysate, SH-SY5Y, C6, mouse liver tissue, human liver tissue, mouse brain tissue.

Subcellular location: Cytoplasm, Endomembrane system, Cytoplasmic vesicle, Microtubule, Membrane.

Database links: SwissProt: Q9H492 Human | Q91VR7 Mouse | Q6XVN8 Rat

Recommended Dilutions:

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

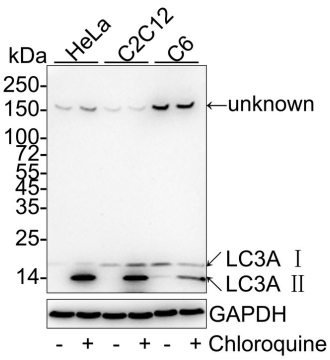

Fig1: Western blot analysis of MAP1LC3A on different lysates with Rabbit anti-MAP1LC3A antibody (ET1609-26) at 1/1,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HeLa treated with 50μM Chloroquine for 18 hours cell lysate
Lane 3: C2C12 cell lysate
Lane 4: C2C12 treated with 50μM Chloroquine for 18 hours cell lysate
Lane 5: C6 cell lysate
Lane 6: C6 treated with 50μM Chloroquine for 18 hours cell lysate

Lysates/proteins at 20 μg/Lane.

Predicted band size: 14 kDa
Observed band size: 14/16 kDa

Exposure time: 3 minutes; 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 (ET1609-26) 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.

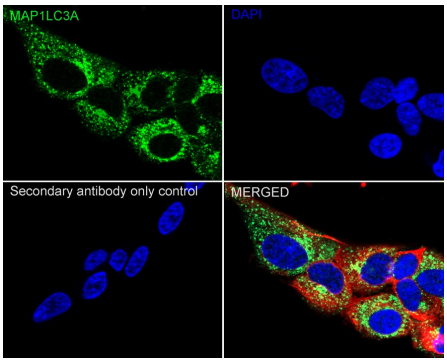

Fig2: Immunocytochemistry analysis of SH-SY5Y cells labeling MAP1LC3A with Rabbit anti-MAP1LC3A antibody (ET1609-26) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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-MAP1LC3A antibody (ET1609-26) 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 (HA601187, 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.

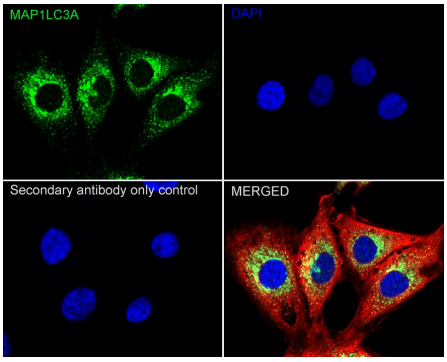

Fig3: Immunocytochemistry analysis of C6 cells labeling MAP1LC3A with Rabbit anti-MAP1LC3A antibody (ET1609-26) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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-MAP1LC3A antibody (ET1609-26) 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 (HA601187, 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.

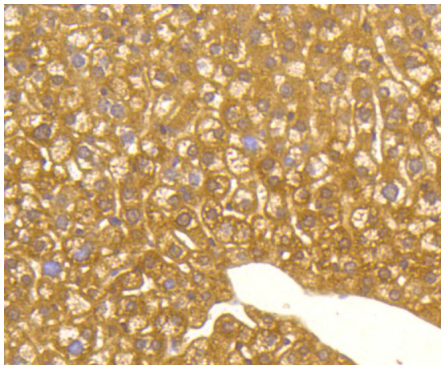

Fig4: Immunohistochemical analysis of paraffin-embedded mouse liver tissue using anti-MAP1LC3A antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1609-26, 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.

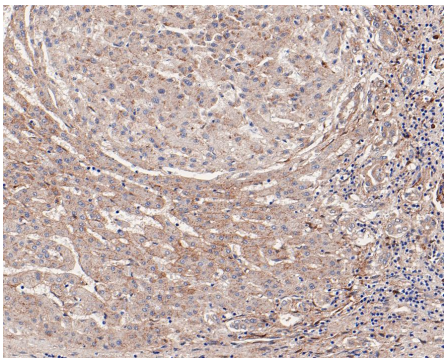

Fig5: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-MAP1LC3A antibody (ET1609-26) at 1/50 dilution.

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 (ET1609-26) at 1/50 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.

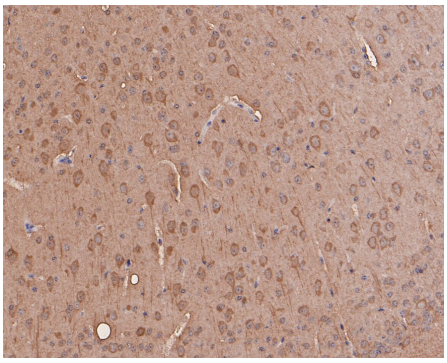

Fig6: Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Rabbit anti-MAP1LC3A antibody (ET1609-26) at 1/200 dilution.

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 (ET1609-26) 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.

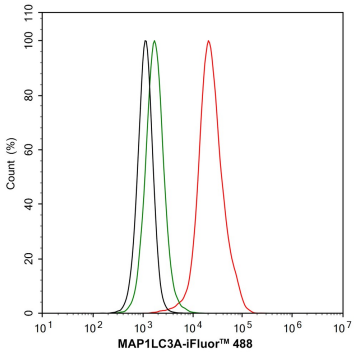

Fig7: Flow cytometric analysis of SH-SY5Y cells labeling MAP1LC3A.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1609-26, 1/1,000) (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).

Background References

1. Kim JY et al. Expression of autophagy-related proteins according to androgen receptor and HER-2 status in estrogen receptor-negative breast cancer. PLoS One 9:e105666 (2014).
2. Jung CH et al. ULK-Atg13-FIP200 complexes mediate mTOR signaling to the autophagy machinery. Mol Biol Cell 20:1992-2003 (2009).