

ATG16L Antibody

Purified Rabbit Polyclonal Antibody (Pab) Catalog # AP1817d

Specification

ATG16L Antibody - Product Information

Application Primary Accession Reactivity Host Clonality Isotype WB, IF,E <u>Q676U5</u> Human Rabbit Polyclonal Rabbit IgG

ATG16L Antibody - Additional Information

Gene ID 55054

Other Names Autophagy-related protein 16-1, APG16-like 1, ATG16L1, APG16L

Target/Specificity

This APG16L antibody is generated from rabbits immunized with a recombinant fragment protein from human APG16L.

Dilution WB~~1:1000 IF~~1:100 E~~Use at an assay dependent concentration.

Format

Purified polyclonal antibody supplied in PBS with 0.09% (W/V) sodium azide. This antibody is prepared by Saturated Ammonium Sulfate (SAS) precipitation followed by dialysis against PBS.

Storage

Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.

Precautions ATG16L Antibody is for research use only and not for use in diagnostic or therapeutic procedures.

ATG16L Antibody - Protein Information

Name ATG16L1 {ECO:0000303|PubMed:17200669, ECO:0000312|HGNC:HGNC:21498}

Function Plays an essential role in both canonical and non-canonical autophagy: interacts with ATG12-ATG5 to mediate the lipidation to ATG8 family proteins (MAP1LC3A, MAP1LC3B, MAP1LC3C, GABARAPL1, GABARAPL2 and GABARAP) (PubMed:23376921, PubMed:23392225, PubMed:24553140, PubMed:24954904, PubMed:27273576, PubMed:29317426,



PubMed: 30778222, PubMed: 33909989). Acts as a molecular hub, coordinating autophagy pathways via distinct domains that support either canonical or non- canonical signaling (PubMed:29317426, PubMed:30778222). During canonical autophagy, interacts with ATG12-ATG5 to mediate the conjugation of phosphatidylethanolamine (PE) to ATG8 proteins, to produce a membrane-bound activated form of ATG8 (PubMed:23376921, PubMed:23392225, PubMed:24553140, PubMed:24954904, PubMed:27273576). Thereby, controls the elongation of the nascent autophagosomal membrane (PubMed:23376921, PubMed:23392225, PubMed:24553140, PubMed:24954904, PubMed:27273576). As part of the ATG8 conjugation system with ATG5 and ATG12, required for recruitment of LRRK2 to stressed lysosomes and induction of LRRK2 kinase activity in response to lysosomal stress (By similarity). Also involved in non-canonical autophagy, a parallel pathway involving conjugation of ATG8 proteins to single membranes at endolysosomal compartments, probably by catalyzing conjugation of phosphatidylserine (PS) to ATG8 (PubMed:<u>33909989</u>). Non-canonical autophagy plays a key role in epithelial cells to limit lethal infection by influenza A (IAV) virus (By similarity). Regulates mitochondrial antiviral signaling (MAVS)-dependent type I interferon (IFN-I) production (PubMed:22749352, PubMed:25645662). Negatively regulates NOD1- and NOD2-driven inflammatory cytokine response (PubMed:24238340). Instead, promotes an autophagy-dependent antibacterial pathway together with NOD1 or NOD2 (PubMed: 20637199). Plays a role in regulating morphology and function of Paneth cell (PubMed:<u>18849966</u>).

Cellular Location

Cytoplasm. Preautophagosomal structure membrane; Peripheral membrane protein. Endosome membrane; Peripheral membrane protein. Lysosome membrane; Peripheral membrane protein. Note=Recruited to omegasomes membranes by WIPI2 (By similarity). Omegasomes are endoplasmic reticulum connected strutures at the origin of preautophagosomal structures (By similarity). Localized to preautophagosomal structure (PAS) where it is involved in the membrane targeting of ATG5 (By similarity). Also localizes to discrete punctae along the ciliary axoneme (By similarity). Upon activation of non-canonical autophagy, recruited to single-membrane endolysosomal compartments (PubMed:29317426). Under starved conditions, the ATG12-ATG5-ATG16L1 complex is translocated to phagophores driven by RAB33B (PubMed:32960676). {ECO:0000250|UniProtKB:Q8C0J2, ECO:0000269|PubMed:29317426, ECO:0000269|PubMed:32960676}

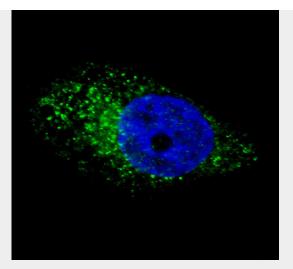
ATG16L Antibody - Protocols

Provided below are standard protocols that you may find useful for product applications.

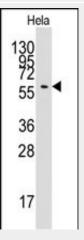
- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

ATG16L Antibody - Images





Fluorescent image of U251 cells stained with ATG16L antibody. U251 cells were treated with Chloroquine (50 μ M,16h), then fixed with 4% PFA (20 min), permeabilized with Triton X-100 (0.2%, 30 min). Cells were then incubated with AP1817d ATG16L primary antibody (1:100, 2 h at room temperature). For secondary antibody, Alexa Fluor® 488 conjugated donkey anti-rabbit antibody (green) was used (1:1000, 1h). Nuclei were counterstained with Hoechst 33342 (blue) (10 μ g/ml, 5 min). ATG16L immunoreactivity is localized to autophagic vacuoles in the cytoplasm of U251 cells, supported by Human Protein Atlas Data (http://www.proteinatlas.org/ENSG0000085978).



Western blot analysis of anti-APG16 Pab (Cat.#AP1817d) in Hela cell line lysates (35ug/lane).APG16(arrow) was detected using the purified Pab.

ATG16L Antibody - Background

Macroautophagy is the major inducible pathway for the general turnover of cytoplasmic constituents in eukaryotic cells, it is also responsible for the degradation of active cytoplasmic enzymes and organelles during nutrient starvation. Macroautophagy involves the formation of double-membrane bound autophagosomes which enclose the cytoplasmic constituent targeted for degradation in a membrane bound structure, which then fuse with the lysosome (or vacuole) releasing a single-membrane bound autophagic bodies which are then degraded within the lysosome (or vacuole).

The APG12-APG5-APG16L complex is esential for the elongation of autophagic isolation membranes. This complex initially associates in uniform distribution with small vesicle membranes. During membrane elongation, the complex partitions, with a great concentration building on the outer side of the isolation membrane. Upon completion of the formation of the autophagosome, the APG12-APG5-APG16L dissociates from the membrane.



ATG16L Antibody - References

References for protein:

1.Baehrecke EH. Nat Rev Mol Cell Biol. 6(6):505-10. (2005)

2.Lum JJ, et al. Nat Rev Mol Cell Biol. 6(6):439-48. (2005)

3.Greenberg JT. Dev Cell. 8(6):799-801. (2005)

4.Levine B. Cell. 120(2):159-62. (2005)

5.Shintani T and Klionsky DJ. Science. 306(5698):990-5. (2004)

References for U251 cell line:

1. Westermark B.; Pontén J.; Hugosson R. (1973)." Determinants for the establishment of permanent tissue culture lines from human gliomas". Acta Pathol Microbiol Scand A. 81:791-805. [PMID: 4359449].

2. Pontén, J., Westermark B. (1978)." Properties of Human Malignant Glioma Cells in Vitro". Medical Biology 56: 184-193.[PMID: 359950].

3. Geng Y.;Kohli L.; Klocke B.J.; Roth K.A.(2010). "Chloroquine-induced autophagic vacuole accumulation and cell death in glioma cells is p53 independent". Neuro Oncol. 12(5): 473–481.[PMID: 20406898].

ATG16L Antibody - Citations

• <u>Autophagy in cancer associated fibroblasts promotes tumor cell survival: Role of hypoxia.</u> <u>HIF1 induction and NFκB activation in the tumor stromal microenvironment.</u>