

APG5 Antibody

Purified Rabbit Polyclonal Antibody (Pab) Catalog # AP1812C

Specification

APG5 Antibody - Product Information

Application Primary Accession Reactivity Host Clonality Isotype

WB, IF, IHC-P,E <u>Q9H1Y0</u> Human Rabbit Polyclonal Rabbit IgG

APG5 Antibody - Additional Information

Gene ID 9474

Other Names Autophagy protein 5, APG5-like, Apoptosis-specific protein, ATG5, APG5L, ASP

Target/Specificity

This ATG5 antibody is generated from rabbits immunized with a KLH conjugated synthetic peptide selected from the full length of human ATG5.

Dilution WB~~1:500 IF~~1:200 IHC-P~~1:10~50 E~~Use at an assay dependent concentration.

Format

Purified monoclonal antibody supplied in PBS with 0.09% (W/V) sodium azide. This antibody is purified through a protein A column, 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 APG5 Antibody is for research use only and not for use in diagnostic or therapeutic procedures.

APG5 Antibody - Protein Information

Name ATG5 (<u>HGNC:589</u>)

Synonyms APG5L, ASP

Function Involved in autophagic vesicle formation. Conjugation with ATG12, through a



ubiquitin-like conjugating system involving ATG7 as an E1-like activating enzyme and ATG10 as an E2-like conjugating enzyme, is essential for its function. The ATG12-ATG5 conjugate acts as an E3-like enzyme which is required for lipidation of ATG8 family proteins and their association to the vesicle membranes. Involved in mitochondrial quality control after oxidative damage, and in subsequent cellular longevity. Plays a critical role in multiple aspects of lymphocyte development and is essential for both B and T lymphocyte survival and proliferation. Required for optimal processing and presentation of antigens for MHC II. Involved in the maintenance of axon morphology and membrane structures, as well as in normal adipocyte differentiation. Promotes primary ciliogenesis through removal of OFD1 from centriolar satellites and degradation of IFT20 via the autophagic pathway. As part of the ATG8 conjugation system with ATG12 and ATG16L1, required for recruitment of LRRK2 to stressed lysosomes and induction of LRRK2 kinase activity in response to lysosomal stress (By similarity).

Cellular Location

Cytoplasm. Preautophagosomal structure membrane; Peripheral membrane protein. Note=Colocalizes with nonmuscle actin. The conjugate detaches from the membrane immediately before or after autophagosome formation is completed (By similarity). Also localizes to discrete punctae along the ciliary axoneme and to the base of the ciliary axoneme. Under starved conditions, the ATG12-ATG5-ATG16L1 complex is translocated to phagophores driven by RAB33B (PubMed:32960676). {ECO:0000250, ECO:0000269|PubMed:32960676}

Tissue Location

Ubiquitous. The mRNA is present at similar levels in viable and apoptotic cells, whereas the protein is dramatically highly expressed in apoptotic cells

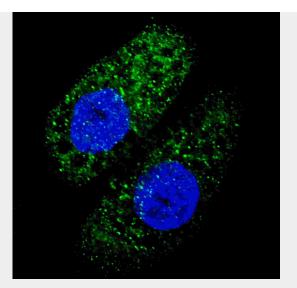
APG5 Antibody - Protocols

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

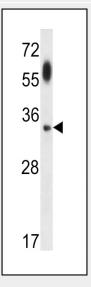
- <u>Western Blot</u>
- <u>Blocking Peptides</u>
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

APG5 Antibody - Images



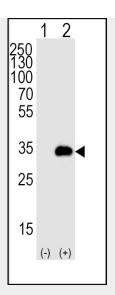


Fluorescent image of U251 cells stained with ATG5 antibody. U251 cells were treated with Chloroquine (50 μ M,16h), then fixed with 4% PFA (20 min), permeabilized with Triton X-100 (0.2%, 30 min). Cells were then incubated with AP1812c ATG5 primary antibody (1:200, 2 h at room temperature). For secondary 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). ATG5 immunoreactivity is localized to autophagic vacuoles in the cytoplasm of U251 cells.

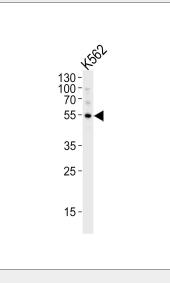


ATG5 Antibody (Cat.#AP1812c) western blot analysis in uterus tumor cell line lysates (35ug/lane).This demonstrates the ATG5 antibody detected the ATG5 protein (arrow).

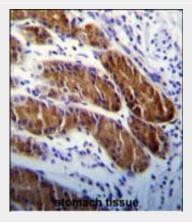




Western blot analysis of APG5 (arrow) using rabbit polyclonal APG5 Antibody (Cat. #AP1812c). 293T cell lysates either nontransfected (Lane 1) or transiently transfected (Lane 2) with the APG5 gene.



Western blot analysis of lysate from K562 cell line, using APG5 Antibody(Cat. #AP1812c). AP1812c was diluted at 1:1000. A goat anti-rabbit IgG H&L(HRP) at 1:10000 dilution was used as the secondary antibody. Lysate at 20ug.



ATG5 Antibody (Cat. #AP1812c)immunohistochemistry analysis in formalin fixed and paraffin



embedded human stomach tissue followed by peroxidase conjugation of the secondary antibody and DAB staining. This data demonstrates the use of ATG5 Antibody for immunohistochemistry. Clinical relevance has not been evaluated.

APG5 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). APG5, required for autophagy, conjugates to ATG12 and associates with an isolation membrane to form a cup-shaped isolation membrane and autophagosome. The conjugate detaches from the membrane immediately before or after autophagosome formation is completed. APG5 may also play an important role in the apoptotic process, possibly within the modified cytoskeleton. Its expression is a relatively late event in the apoptotic process, occurring downstream of caspase activity.

APG5 Antibody - References

References for protein:

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

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7.Strausberg R.L., et al. PNAS 99:16899-16903(2002)

8.Grand R.J.A., et al. Exp. Cell Res. 218:439-451(1995)

9. Mizushima N., et al. J. Biol. Chem. 273:33889-33892(1998)

10. Mizushima N., et al. J. Cell Biol. 152:657-668(2001)

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].

APG5 Antibody - Citations

• <u>Class III PI3K Vps34 plays an essential role in autophagy and in heart and liver function.</u>