

**ATG4D Antibody (N-term)**  
**Purified Rabbit Polyclonal Antibody (Pab)**  
**Catalog # AP11448a****Specification**

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**ATG4D Antibody (N-term) - Product Information**

Application	IF, WB, IHC-P,E
Primary Accession	<a href="#">Q86TLO</a>
Other Accession	<a href="#">NP_116274.3</a>
Reactivity	Mouse
Host	Rabbit
Clonality	Polyclonal
Isotype	Rabbit IgG
Calculated MW	52922
Antigen Region	42-71

**ATG4D Antibody (N-term) - Additional Information****Gene ID** 84971**Other Names**

Cysteine protease ATG4D, 3422-, AUT-like 4 cysteine endopeptidase, Autophagin-4, Autophagy-related cysteine endopeptidase 4, Autophagy-related protein 4 homolog D, Cysteine protease ATG4D, mitochondrial, ATG4D, APG4D, AUTL4

**Target/Specificity**

This ATG4D antibody is generated from rabbits immunized with a KLH conjugated synthetic peptide between 42-71 amino acids from the N-terminal region of human ATG4D.

**Dilution**

IF~~1:200  
WB~~1:1000  
IHC-P~~1:50~100

**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**

ATG4D Antibody (N-term) is for research use only and not for use in diagnostic or therapeutic procedures.

**ATG4D Antibody (N-term) - Protein Information**

**Name** ATG4D {ECO:0000303|PubMed:19549685, ECO:0000312|HGNC:HGNC:20789}

**Function** [Cysteine protease ATG4D]: Cysteine protease that plays a key role in autophagy by mediating both proteolytic activation and delipidation of ATG8 family proteins (PubMed:[21177865](#), PubMed:[29458288](#), PubMed:[30661429](#)). The protease activity is required for proteolytic activation of ATG8 family proteins: cleaves the C-terminal amino acid of ATG8 proteins MAP1LC3 and GABARAPL2, to reveal a C-terminal glycine (PubMed:[21177865](#)). Exposure of the glycine at the C-terminus is essential for ATG8 proteins conjugation to phosphatidylethanolamine (PE) and insertion to membranes, which is necessary for autophagy (By similarity). In addition to the protease activity, also mediates delipidation of ATG8 family proteins (PubMed:[29458288](#), PubMed:[33909989](#)). Catalyzes delipidation of PE-conjugated forms of ATG8 proteins during macroautophagy (PubMed:[29458288](#), PubMed:[33909989](#)). Also involved in non-canonical autophagy, a parallel pathway involving conjugation of ATG8 proteins to single membranes at endolysosomal compartments, by catalyzing delipidation of ATG8 proteins conjugated to phosphatidylserine (PS) (PubMed:[33909989](#)). ATG4D plays a role in the autophagy-mediated neuronal homeostasis in the central nervous system (By similarity). Compared to other members of the family (ATG4A, ATG4B or ATG4C), constitutes the major protein for the delipidation activity, while it promotes weak proteolytic activation of ATG8 proteins (By similarity). Involved in phagophore growth during mitophagy independently of its protease activity and of ATG8 proteins: acts by regulating ATG9A trafficking to mitochondria and promoting phagophore- endoplasmic reticulum contacts during the lipid transfer phase of mitophagy (PubMed:[33773106](#)).

#### **Cellular Location**

[Cysteine protease ATG4D]: Cytoplasm

#### **Tissue Location**

Widely expressed in testis.

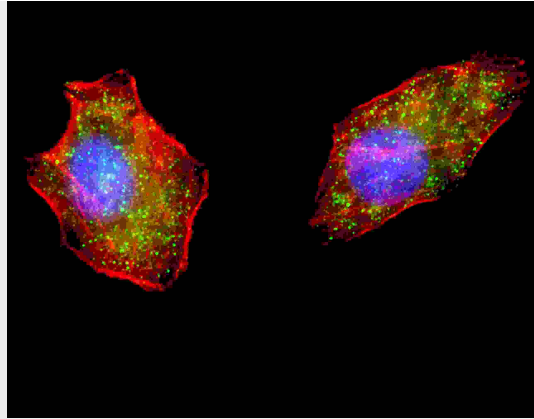
#### **ATG4D Antibody (N-term) - Protocols**

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

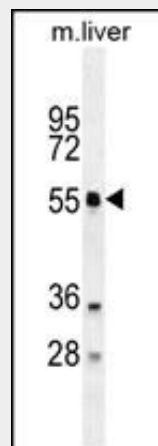
- [Western Blot](#)
- [Blocking Peptides](#)
- [Dot Blot](#)
- [Immunohistochemistry](#)
- [Immunofluorescence](#)
- [Immunoprecipitation](#)
- [Flow Cytometry](#)
- [Cell Culture](#)

#### **ATG4D Antibody (N-term) - Images**

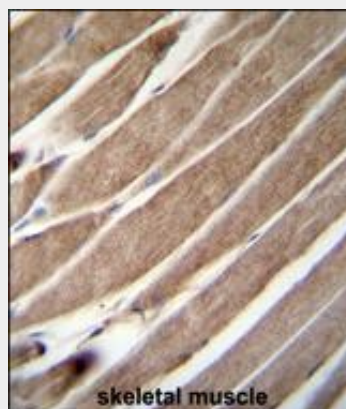




Fluorescent image of U251 cells stained with ATG4D (N-term) antibody. U251 cells were treated with Chloroquine (50  $\mu$ M, 16h), then fixed with 4% PFA (20 min), permeabilized with Triton X-100 (0.2%, 30 min). Cells were then incubated with AP11448a ATG4D (N-term) primary antibody (1:200, 2 h at room temperature). For secondary antibody, Alexa Fluor® 488 conjugated donkey anti-rabbit antibody (green) was used (1:1000, 1h). Cytoplasmic actin was counterstained with Alexa Fluor® 555 (red) conjugated Phalloidin (5.25  $\mu$ M, 25 min). Nuclei were counterstained with Hoechst 33342 (blue) (10  $\mu$ g/ml, 5 min). Pictures were taken on a Bioevo microscope (BZ-900, Keyence). ATG4D (N-term) immunoreactivity is localized to autophagic vacuoles in the cytoplasm of U251 cells.



ATG4D Antibody (N-term) (Cat. #AP11448a) western blot analysis in mouse liver tissue lysates (35ug/lane). This demonstrates the ATG4D antibody detected the ATG4D protein (arrow).



ATG4D Antibody (N-term) (Cat. #AP11448a) immunohistochemistry analysis in formalin fixed and paraffin embedded human skeletal muscle followed by peroxidase conjugation of the secondary

antibody and DAB staining. This data demonstrates the use of ATG4D Antibody (N-term) for immunohistochemistry. Clinical relevance has not been evaluated.

### **ATG4D Antibody (N-term) - Background**

Autophagy is the process by which endogenous proteins and damaged organelles are destroyed intracellularly. Autophagy is postulated to be essential for cell homeostasis and cell remodeling during differentiation, metamorphosis, non-apoptotic cell death, and aging. Reduced levels of autophagy have been described in some malignant tumors, and a role for autophagy in controlling the unregulated cell growth linked to cancer has been proposed. This gene encodes a member of the autophagin protein family. The encoded protein is also designated as a member of the C-54 family of cysteine proteases.

### **ATG4D Antibody (N-term) - References**

References for protein:

1. Betin, V.M., et al. *J. Cell. Sci.* 122 (PT 14), 2554-2566 (2009)
2. Lamesch, P., et al. *Genomics* 89(3):307-315(2007)
3. Marino, G., et al. *J. Biol. Chem.* 278(6):3671-3678(2003)

References for U251 cell line:

1. Westermarck 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., Westermarck 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].