

# Phospho-ULK1(S556) Antibody

Affinity Purified Rabbit Polyclonal Antibody (Pab) Catalog # AP3804a

# **Specification**

# Phospho-ULK1(S556) Antibody - Product Information

**Application** DB, IF,E **Primary Accession** 075385 NP 003556.1 Other Accession Reactivity Human Host **Rabbit** Clonality **Polyclonal** Isotype Rabbit IgG Calculated MW 112631

## Phospho-ULK1(S556) Antibody - Additional Information

#### **Gene ID 8408**

### **Other Names**

Serine/threonine-protein kinase ULK1, Autophagy-related protein 1 homolog, ATG1, hATG1, Unc-51-like kinase 1, ULK1, KIAA0722

#### Target/Specificity

This ULK1 Antibody is generated from rabbits immunized with a KLH conjugated synthetic phosphopeptide corresponding to amino acid residues surrounding S556 of human ULK1.

# **Dilution**

DB~~1:500 IF~~1:200

E~~Use at an assay dependent concentration.

#### Format

Purified polyclonal antibody supplied in PBS with 0.09% (W/V) sodium azide. This antibody is purified through a protein A column, followed by peptide affinity purification.

## **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**

Phospho-ULK1(S556) Antibody is for research use only and not for use in diagnostic or therapeutic procedures.

# Phospho-ULK1(S556) Antibody - Protein Information

Name ULK1 {ECO:0000303|PubMed:9693035, ECO:0000312|HGNC:HGNC:12558}



Function Serine/threonine-protein kinase involved in autophagy in response to starvation (PubMed:18936157, PubMed:21460634, PubMed:21795849, PubMed:23524951, PubMed:25040165, PubMed:29487085, PubMed:31123703). Acts upstream of phosphatidylinositol 3-kinase PIK3C3 to regulate the formation of autophagophores, the precursors of autophagosomes (PubMed: <u>18936157</u>, PubMed: <u>21460634</u>, PubMed: <u>21795849</u>, PubMed: <u>25040165</u>). Part of regulatory feedback loops in autophagy: acts both as a downstream effector and negative regulator of mammalian target of rapamycin complex 1 (mTORC1) via interaction with RPTOR (PubMed: 21795849). Activated via phosphorylation by AMPK and also acts as a regulator of AMPK by mediating phosphorylation of AMPK subunits PRKAA1, PRKAB2 and PRKAG1, leading to negatively regulate AMPK activity (PubMed: 21460634). May phosphorylate ATG13/KIAA0652 and RPTOR; however such data need additional evidences (PubMed: 18936157). Plays a role early in neuronal differentiation and is required for granule cell axon formation (PubMed:11146101). Also phosphorylates SESN2 and SQSTM1 to regulate autophagy (PubMed: 25040165, PubMed:37306101). Phosphorylates FLCN, promoting autophagy (PubMed:25126726). Phosphorylates AMBRA1 in response to autophagy induction, releasing AMBRA1 from the cytoskeletal docking site to induce autophagosome nucleation (PubMed: 20921139). Phosphorylates ATG4B, leading to inhibit autophagy by decreasing both proteolytic activation and delipidation activities of ATG4B (PubMed: 28821708).

#### **Cellular Location**

Cytoplasm, cytosol. Preautophagosomal structure. Note=Under starvation conditions, is localized to puncate structures primarily representing the isolation membrane that sequesters a portion of the cytoplasm resulting in the formation of an autophagosome.

#### **Tissue Location**

Ubiquitously expressed. Detected in the following adult tissues: skeletal muscle, heart, pancreas, brain, placenta, liver, kidney, and lung

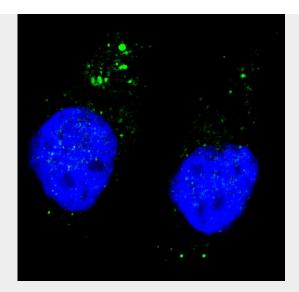
# Phospho-ULK1(S556) Antibody - Protocols

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

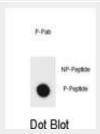
- Western Blot
- Blocking Peptides
- Dot Blot
- <u>Immunohistochemistry</u>
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

# Phospho-ULK1(S556) Antibody - Images





Fluorescent image of U251 cells stained with ULK1 (phospho S556) 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 AP3804a ULK1 (phospho S556) 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  $\mu$ g/ml, 5 min). ULK1 (phospho S556) immunoreactivity is localized to autophagic vacuoles in the cytoplasm of U251 cells.



Dot blot analysis of ULK1 Antibody (Phospho S556) Phospho-specific Pab (Cat. #AP3804a) on nitrocellulose membrane. 50ng of Phospho-peptide or Non Phospho-peptide per dot were adsorbed. Antibody working concentrations are 0.6ug per ml.

# Phospho-ULK1(S556) Antibody - Background

Involved in autophagy. Required for autophagosome formation (By similarity). Target of the TOR kinase signaling pathway that regulates autophagy through the control of phosphorylation status of ATG13/KIAA0652 and ULK1, and the regulation of the ATG13-ULK1-RB1CC1 complex (By similarity). Phosphorylates ATG13/KIAA0652. Involved in axon growth (By similarity). Plays an essential role in neurite extension of cerebellar granule cells (By similarity).

# Phospho-ULK1(S556) Antibody - References

# References for protein:

- 1.Mercer, C.A., et al. Autophagy 5(5):649-662(2009)
- 2.Ganley, I.G., et al. J. Biol. Chem. 284(18):12297-12305(2009)
- 3.Jung, C.H., et al. Mol. Biol. Cell 20(7):1992-2003(2009)
- 4. Hosokawa, N., et al. Mol. Biol. Cell 20(7):1981-1991(2009)
- 5.Chan, E.Y. Sci Signal 2 (84), PE51 (2009)

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.





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[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].