# SANTA CRUZ BIOTECHNOLOGY, INC.

# Atg3 (D-9): sc-393623



## BACKGROUND

Atg3 (autophagy-related protein 3), also known as APG3-like, hAPG3 or PC3-96, is an E2-like enzyme that localizes to the cytoplasm and is expressed in a variety of tissues with predominant levels found in kidney, placenta, liver, heart and skeletal muscle. Atg3 catalyzes the formation of the Atg8-phosphatidylethanolamine (Atg8-PE) conjugate, a reaction that is essential for autophagy (a cellular process that allows for the degradation of organelles and bulk cellular proteins). The process of forming the Atg8-PE conjugate begins with the removal of the C-terminal arginine residue of Atg8 by Atg4, a cysteine protease. The, now exposed, glycine residue is then activated by Atg7 and is then transferred to Atg3 for the final conjugation to PE. This last step can be accelerated by the presence of the Atg12-Atg5 conjugate which functions similarly to an E3 enzyme.

#### REFERENCES

- 1. Tanida, I., et al. 2002. Human Apg3p/Aut1p homologue is an authentic E2 enzyme for multiple substrates, GATE-16, GABARAP, and MAP-LC3, and facilitates the conjugation of hApg12p to hApg5p. J. Biol. Chem. 277: 13739-13744.
- Wu, B.X., et al. 2006. The rat Apg3p/Aut1p homolog is upregulated by ischemic preconditioning in the retina. Mol. Vis. 12: 1292-1302.
- Yamada, Y., et al. 2006. Crystallization and preliminary X-ray analysis of Atg3. Acta Crystallogr. Sect. F Struct. Biol. Cryst. Commun. 62: 1016-1017.
- Yamada, Y., et al. 2007. The crystal structure of Atg3, an autophagy-related ubiquitin carrier protein (E2) enzyme that mediates Atg8 lipidation. J. Biol. Chem. 282: 8036-8043.
- Hanada, T., et al. 2007. The Atg12-Atg5 conjugate has a novel E3-like activity for protein lipidation in autophagy. J. Biol. Chem. 282: 37298-37302.
- Shao, Y., et al. 2007. Stimulation of Atg12-Atg5 conjugation by ribonucleic acid. Autophagy 3: 10-16.
- Ma, J., et al. 2007. Overexpression of autophagy-related genes inhibits yeast filamentous growth. Autophagy 3: 604-609.
- Tanida, I., et al. 2008. Consideration about negative controls for LC3 and expression vectors for four colored fluorescent protein-LC3 negative controls. Autophagy 4: 131-134.

#### **CHROMOSOMAL LOCATION**

Genetic locus: ATG3 (human) mapping to 3q13.2; Atg3 (mouse) mapping to 16 B5.

## SOURCE

Atg3 (D-9) is a mouse monoclonal antibody raised against amino acids 1-300 mapping at the N-terminus of Atg3 of human origin.

## PRODUCT

Each vial contains 200  $\mu g \; lgG_1$  kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

#### **APPLICATIONS**

Atg3 (D-9) is recommended for detection of Atg3 of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2  $\mu$ g per 100-500  $\mu$ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Suitable for use as control antibody for Atg3 siRNA (h): sc-72582, Atg3 siRNA (m): sc-72583, Atg3 shRNA Plasmid (h): sc-72582-SH, Atg3 shRNA Plasmid (m): sc-72583-SH, Atg3 shRNA (h) Lentiviral Particles: sc-72582-V and Atg3 shRNA (m) Lentiviral Particles: sc-72583-V.

Molecular Weight of Atg3: 42 kDa.

Positive Controls: Jurkat whole cell lysate: sc-2204, K-562 whole cell lysate: sc-2203 or NAMALWA cell lysate: sc-2234.

## **RECOMMENDED SUPPORT REAGENTS**

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG KBP-HRP: sc-516102 or m-IgG KBP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-IgG KBP-FITC: sc-516140 or m-IgG KBP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

#### DATA





Atg3 (D-9): sc-393623. Western blot analysis of Atg3 expression in Jurkat (A), NIH/3T3 (B), RAW 264.7 (C), PC-12 (D) and KNRK (E) whole cell lysates and mouse liver tissue extract (F).

Atg3 (D-9): sc-393623. Western blot analysis of Atg3 expression in K-562 (A), HEL 92.1.7 (B), NAMALWA (C), WEHI-231 (D) and WR19L (E) whole cell lysates.

#### SELECT PRODUCT CITATIONS

 Park, H.J. and Kim, M.M. 2019. Amentoflavone induces autophagy and modulates p53. Cell J. 21: 27-34.

## **STORAGE**

Store at 4° C, \*\*D0 NOT FREEZE\*\*. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

## **RESEARCH USE**

For research use only, not for use in diagnostic procedures.