

ProductInformation

Anti-Atg3 (C-terminal)

produced in rabbit, IgG fraction of antiserum

Catalog Number **A3606**

Product Description

Anti-Atg3 (C-terminal) is produced in rabbit using as immunogen a synthetic peptide corresponding to amino acids 298-314 of human Atg3 (Gene ID: 64422), conjugated to KLH via a N-terminal cysteine residue. The corresponding sequence is identical in rat and mouse. Whole serum is fractionated and further purified by anion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti-Atg3 (C-terminal) recognizes human, rat, and mouse Atg3 by immunoprecipitation (~36 kDa) and immunohistochemistry. Detection of the Atg3 staining by immunohistochemistry is specifically inhibited with the immunizing peptide.

Macroautophagy, usually referred to as autophagy, is a major pathway for bulk degradation of cytoplasmic constituents and organelles. In this process, portions of the cytoplasm are sequestered into double membrane vesicles, the autophagosomes, and subsequently delivered to the lysosome for degradation and recycling.^{1,2} Although autophagy is a constitutive cellular event, it is enhanced under certain conditions such as starvation, hormonal stimulation and drug treatments.³ Autophagy is required for normal turnover of cellular components during starvation. It plays an essential role in cellular differentiation, cell death and aging. Defective autophagy may contribute to certain human diseases such as cancer, neurodegenerative diseases, muscular disorders and pathogen infections.^{4,5} Autophagy is an evolutionarily conserved pathway seen in all eukaryotic cells.¹ At least 16 ATG genes required for autophagosome formation were identified in yeast by genetic screens. For many of these genes, related homologs have been identified in mammals.⁶

Two ubiquitin-like conjugation systems are involved in autophagosome formation: Atg12 and Atg8 conjugation systems. Atg3, the mammalian homolog of yeast Apg3p/Aut1p, is an E2-like enzyme that catalyzes the conjugation reaction between Atg8 and phosphatidylethanolamine (PE). The Atg8-PE conjugate is essential for autophagosome formation.^{7,8} The ubiquitin-like proteins Atg8 and Atg12 are activated by

Atg7, an E1-like enzyme essential for both conjugation systems. Atg8 is then transferred to Atg3 and conjugated to PE, whereas Atg12 is transferred to Atg10, another E2-like enzyme, followed by conjugation to Atg5.^{9,10} Atg3 interacts with Atg7 to form an E1-E2 complex, and with Atg12, which is a substrate for Atg7 but not for Atg3. Moreover, over-expression of Atg3 facilitates the formation of the Atg12-Atg5 conjugate. Enhanced levels of this conjugate promote the recruitment of the lipidated form of MAP-LC3 onto autophagosomal membranes. Although all three Atg8 mammalian homologs, GATE-16, GABARAP and MAP-LC3, are substrates for Atg3, MAP-LC3 is the preferred substrate. Atg3 is ubiquitously expressed in human tissues.⁷

Reagent

Supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

Product Profile

Immunoprecipitation: a working amount of 2-4 µL is recommended using rat NRK and mouse 3T3 cell lysates.

Immunohistochemistry: a working dilution of 1:50-1:100 is recommended using biotin/ExtrAvidin®-Peroxidase staining of heat-retrieved formalin-fixed paraffin-embedded human cerebellum sections.

Note: In order to obtain the best results using various techniques and preparations, we recommend determining the optimal working dilutions by titration.

References

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