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# **ProductInformation**

# Anti-Atg4B

produced in rabbit, IgG fraction of antiserum

Catalog Number A2981

## **Product Description**

Anti-Atg4B is developed in rabbit using as immunogen a synthetic peptide corresponding to amino acids 6-22 of human Atg4B (Gene ID: 23192), conjugated to KLH via a C-terminal cysteine residue. The corresponding sequence is identical in 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-Atg4B recognizes human, rat, and mouse Atg4B by immunoblotting (~44 kDa) and immunohistochemistry. Detection of the Atg4B band by immunoblotting 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.<sup>3</sup> 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 evolutionary conserved pathway seen in all eukaryotic cells.1

At least 16 genes encoding for autopahgy (ATG) related proteins that are required for autophagosome formation were identified in yeast by genetic screens. For many of these genes, related homologs have been identified in mammals.<sup>6</sup>

Atg4B, a mammalian orthologue of yeast Atg4, is a cysteine protease responsible for the processing of the mammalian Atg8 homologues: GATE-16, GABARAP, and LC3.7 Atg4B removes the carboxyl-terminal region of proLC3 immediately after its synthesis, generating the soluble LC3-I and exposing a carboxyl-terminal Gly<sup>120</sup>. LC3-I is then modified to a membrane-bound form, LC3-II (a LC3-phospholipid conjugate), by mammalian Atg7 and Atg3, which are E1 and E2-like enzymes, respectively. LC3-II is essential in the formation of autophagosomes. Atg4B also delipidates LC3-II, leading to its release from membranes. 7-9 Four human homologues of yeast Atg4 have been identified: HsAtg4A/autophagin-2, HsAtg4B/autophagin-1, autophagin-3, and autophagin-4. HsAtq4A cleaves GATE-16 most efficiently; whereas, HsAtg4B has a broad specificity for mammalian Atg8 homologues, GATE-16, GABARAP, and LC3. 10 Atg4B is widely expressed in human and rat tissues, being most abundant in the brain. 9,11

## 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 is not recommended. Storage in "frost-free" freezers is also 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**

Immunoblotting: a working antibody dilution of 1:500–1:1,000 is recommended using a whole extract of rat brain.

Immunoblotting: a working antibody dilution of 1:125–1:500 is recommended using a whole extract of eTe cells.

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

<u>Note</u>: 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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