

Product Information

Anti-LC3B antibody, Mouse monoclonal
clone LC3B-6, purified from hybridoma cell culture

Product Number **SAB4200361**

Product Description

Anti-LC3B antibody, Mouse monoclonal (mouse IgG2b isotype) is derived from the hybridoma LC3B-6 produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with a synthetic peptide corresponding to the N-terminal region of human LC3B (GeneID: 81631), conjugated to KLH. The corresponding sequence differs by one amino-acid in rat and mouse. The isotype is determined by ELISA using Mouse Monoclonal Antibody Isotyping Reagents, Product Number ISO2. The antibody is purified from culture supernatant of hybridoma cells grown in a bioreactor.

Anti-LC3B antibody, Mouse monoclonal recognizes human LC3B-I and/or LC3B-II by immunoblotting (~18/16 kDa, respectively) and immunoprecipitation. Detection of the LC3B bands by immunoblotting is specifically inhibited by 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 genes encoding for autophagy (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.⁶

Rat microtubule-associated protein light chain 3 (LC3), the mammalian homolog of yeast Atg8/Apg8/Aut7, is essential in the formation of autophagosomes.⁷ LC3 was first identified in rat as a protein that co-purifies with microtubule-associated protein 1A and 1B from rat brain.³ LC3 exists in cells in two forms. One is cytoplasmic, LC3-I (18 kDa) and the other, LC3-II (16 kDa) is associated with the autophagosome membrane. Following synthesis, the carboxy-terminal region of proLC3 is cleaved by the cysteine protease Atg4, generating the soluble LC3-I and exposing a carboxyl terminal Gly¹²⁰. LC3-I is 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.⁷ The amount of LC3 II correlates with the extent of autophagosome formation.³ Three human orthologs of the rat LC3, named MAP1LC3A/LC3A, MAP1LC3B/LC3B and MAP1LC3C/LC3C, were identified. The human proteins exhibit two forms representing the cytosolic (type I) and the membrane associated (type II) forms. The three human isoforms show different expression patterns in human tissues.⁸

LC3B can be used as a autophagosomal marker.^{3,7}

Reagent

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

Antibody Concentration: ~ 1.0 mg/mL

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 at –20 °C 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 dilution samples should be discarded if not used within 12 hours.

Product Profile

Immunoblotting: a working concentration of 1-2 µg/mL is recommended using whole extracts of HEK-293T cells over-expressing human LC3B.

Immunoprecipitation: a working amount of 10-20 µg is recommended using lysates of human U-87 cells.

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

References

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4. Ali, B.R., et al., *J. Cell Sci.*, **117**, 6401-6412 (2004).
5. Zerial, M., and McBride, H., *Nature Rev. Mol. Cell Biol.*, **2**, 107-117 (2001).
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