Human LC3/MAP1LC3A Antibody

Monoclonal Rat IgG_{2B} Clone # 877005

Catalog Number: MAB8558

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human LC3/MAP1LC3A in direct ELISAs.
Source	Monoclonal Rat IgG_{2B} Clone # 877005
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	*E. coli*-derived recombinant human LC3/MAP1LC3A Accession # Q9H492
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

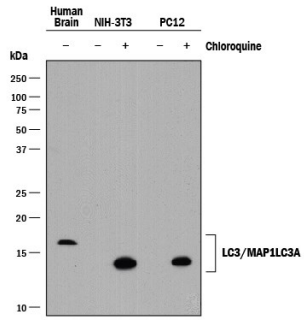
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	2 µg/mL	See Below
Immunohistochemistry	8-25 µg/mL	See Below

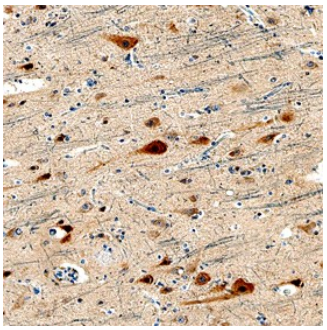
DATA

Western Blot



Detection of Human, Mouse, and Rat LC3/MAP1LC3A by Western Blot. Western blot shows lysates of human brain tissue, NIH-3T3 mouse embryonic fibroblast cell line, and PC-12 rat adrenal pheochromocytoma cell line untreated (-) or treated (+) with 50 µM Chloroquine for 18 hours. PVDF membrane was probed with 2 µg/mL of Rat Anti-Human LC3/MAP1LC3A Monoclonal Antibody (Catalog # MAB8558) followed by HRP-conjugated Anti-Rat IgG Secondary Antibody (Catalog # HAF005). Specific bands were detected for LC3/MAP1LC3A at approximately 14 and 16 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry



LC3/MAP1LC3A in Human Brain Cortex Tissue. LC3/MAP1LC3A was detected in immersion fixed paraffin-embedded sections of human brain cortex tissue using Rat Anti-Human LC3/MAP1LC3A Monoclonal Antibody (Catalog # MAB8558) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Rat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS017) and counterstained with hematoxylin (blue). Specific staining was localized to neurons. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Human Microtubule-associated Protein (MAP) Light Chain 3 (LC3) A is a121 amino acid (aa) protein with a predicted molecular weight of 14 kDa. It is a member of the LC3 subfamily of Autophagy-related 8 (Atg8) proteins (1). The LC3 subfamily also includes LC3B andLC3C. LC3 exhibits 100% aa sequence identity with its mouse and rat orthologs, and is orthologous to the yeast autophagy-related protein Atg8. Atg8 family members show structural similarity with Ubiquitin, but lack aa sequence similarity. LC3 was originally described as part is part of a complex that includes heavy and light chains comprising the MAP1 family of microtubule regulatory proteins (3). However, LC3 has gained attention for MAP1-independent functions in autophagy. LC3 utilizes a ubiquitin-like conjugation system that includes E1-, E2-, and E3-like enzymes to covalently attach phosphatidylethanolamine (PE) to its C-terminus, incorporating it into the phagophore membrane during the early stages of autophagasome formation (4). Recruitment of LC3 to the phagophore may promote membrane elongation (4,5). It may also be involved in cargo recruitment to autophagosomes (1). LC3 is often used as a marker of autophagy.

References:

1. Shpilka, T. *et al.* (2011) Genome Biol. **12**:226.
2. He, H. *et al.* (2003) J. Biol. Chem. **278**:29278.
3. Kuznetsov, S.A. & V.I. Gelfand (1987) FEBS Let. **212**:145.
4. Weidberg, H. *et al.* (2011) Ann Rev. Biochem. **80**:125.
5. Weidberg, H. *et al.* (2010) EMBO J. **29**:1792.