COSMO BIO CO., LTD.
Inspiration for Life Science

MONOCLONAL ANTIBODY

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Catalog No. CTB-LC3-2-IC

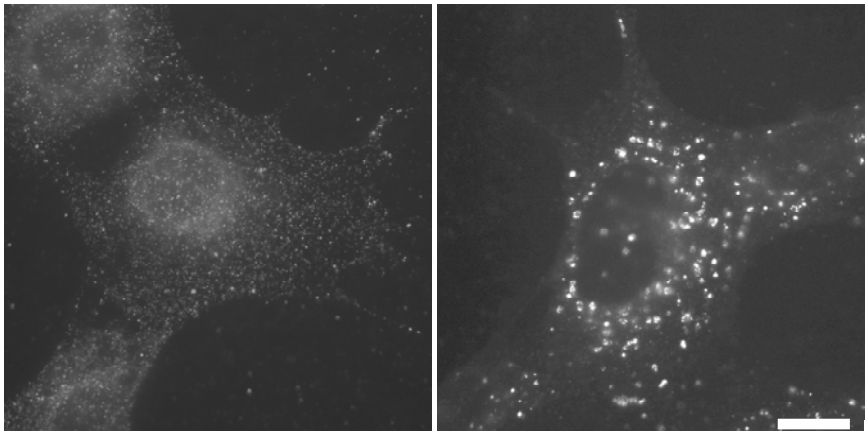
Anti LC3 (Clone: LC3･1703)

BACKGROUND

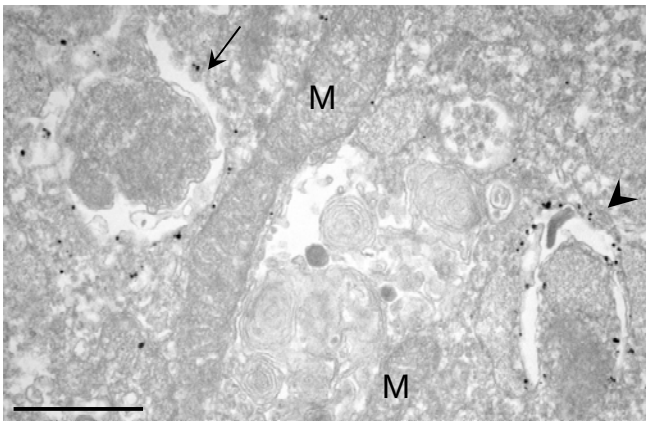
LC3B is one of the mammalian Atg8 homologs and widely used as an autophagosome marker. Immediately after synthesis, LC3 is processed by Atg4 and becomes LC3-I. Upon induction of autophagy, the C-terminal glycine of LC3-I is conjugated to phosphatidylethanolamine, resulting in formation of membrane-bound LC3-II. Most LC3-II is thought be present on autophagosome membrane. The autophagosome subsequently fuses with a lysosome, where inside materials, including LC3-II, are degraded. The expression level of LC3-II generally correlates with the number of autophagosome.

Product type	Primary antibodies
Host	Mouse
Source	
Form	Liquid Protein G purified PBS (pH7.4) with 1% BSA and less than 0.1% NaN_3 as a preservative.
Volume	500 μl
Concentration	0.1mg/ml
Specificity	LC3
Antigen	Human Recombinant LC3
Clone	LC3･1703
Isotype	IgG1

Application notes	ICC, Immuno EM **Recommended use** **Recommended dilutions** ICC: 1/100 Immuno EM : 1/10 Optimal dilutions/concentrations should be determined by the end user. **Staining Pattern**
Cross reactivity	Human
Storage	Store below -20°C (below -70°C for prolonged storage). Aliquot to avoid cycles of freeze/thaw.
References	1) Kabeya, Y., Mizushima, N., Ueno, T., Yamamoto, A., Kirisako, T., Noda, T., Kominami, E., Ohsumi, Y. and Yoshimori, T. LC3, a mammalian homologue of yeast Apg8p, is localized in autophagosome membranes after processing EMBO J. 19, 5720-5728. (2000) 2) Mizushima, N., Yoshimori, T. How to interpret LC3 immunoblotting Autophagy 3:542-545 (2007) 3) Mizushima, N., Yoshimori, T. and Levine, B. Methods in mammalian autophagy research. Cell 140; 313-326 (2010)



(Fig 1) Immunofluorescence microscopy analysis of mouse embryonic fibroblasts (MEFs).
MEFs were cultured in regular DMEM supplemented with 10% FBS (left) or DMEM without amino acids (right) for 1 hr. After fixation with 4% paraformaldehyde for 10 min at room temperature, they were permeabilized with 50μg/ml digitonin for 5 min. Cells were then subjected to immunofluorescence microscopy using #1703 anti-LC3 antibody at 1:100 dilution. Scale bar, 20μm.



(Fig 2) Immuno-electron microscopy analysis of mouse embryonic fibroblasts (MEFs).
MEFs were cultured in DMEM without amino acids for 2 hr. After fixation with 4% paraformaldehyde for 2 hr at room temperature, they were permeabilized with liquid nitrogen. Immuno-electron microscopy analysis (pre-embedding method) of endogenous LC3 was performed using #1703 anti-LC3 antibody at 1:10 dilution. The gold labeling was intensified by using a silver enhancement kit (HQ silver enhancement kit, Nanoprobes, NY). Scale bar, 500 nm.

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