Human/Mouse ATG7 Antibody

Monoclonal Mouse IgG_1 Clone # 683906

Catalog Number: MAB6608

DESCRIPTION

Species Reactivity	Human/Mouse
Specificity	Detects human ATG7 in direct ELISAs and Western blots. In direct ELISAs and Western blots, no cross-reactivity with recombinant human ATG3, 4A, 4B, 5, 10, 12, or 16L1 is observed.
Source	Monoclonal Mouse IgG_1 Clone # 683906
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	*E. coli*-derived recombinant human ATG7 Asn387-Gln570 Accession # O95352
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 either lyophilized or as a 0.2 μm filtered solution in PBS.

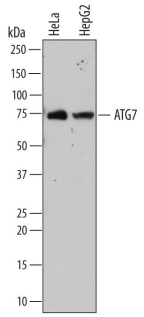
APPLICATIONS

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
Immunocytochemistry	8-25 µg/mL	See Below
Immunohistochemistry	8-25 µg/mL	See Below
Intracellular Staining by Flow Cytometry	0.25 µg/10^6 cells	See Below
Simple Western	20 µg/mL	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

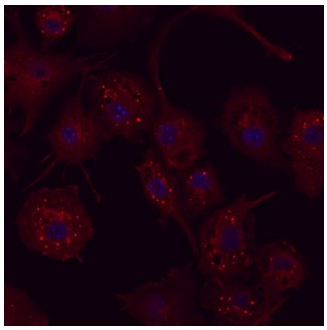
DATA

Western Blot



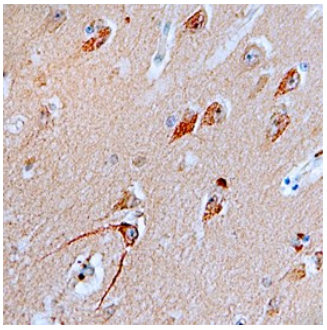
Detection of Human ATG7 by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line and HepG2 human hepatocellular carcinoma cell line. PVDF membrane was probed with 2 µg/mL of Mouse Anti-Human ATG7 Monoclonal Antibody (Catalog # MAB6608) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF007). A specific band was detected for ATG7 at approximately 75 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



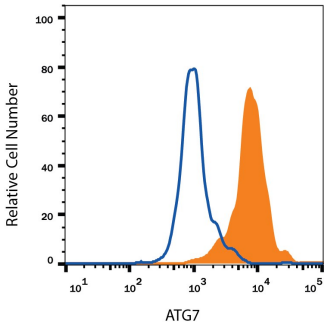
ATG7 in RAW 264.7 Mouse Cell Line. ATG7 was detected in immersion fixed RAW 264.7 mouse monocyte/macrophage cell line using Mouse Anti-Human ATG7 Monoclonal Antibody (Catalog # MAB6608) at 25 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to autophagosomes. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.

Immunohistochemistry



ATG7 in Human Brain. ATG7 was detected in formalin fixed paraffin-embedded sections of human brain (cortex) using Mouse Anti-Human ATG7 Monoclonal Antibody (Catalog # MAB6608) at 15 µg/mL overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using the Anti-Mouse HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS002) and counter-stained with hematoxylin (blue). Specific staining was localized to neuronal cell bodies and processes. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Intracellular Staining by Flow Cytometry



Detection of ATG7 in HeLa Human Cell Line by Flow Cytometry. HeLa human cervical epithelial carcinoma cell line was stained with Mouse Anti-Human/Mouse ATG7 Monoclonal Antibody (Catalog # MAB6608, filled histogram) or isotype control antibody (Catalog # MAB002, open histogram), followed by Allophycocyanin-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0101B). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (Catalog # FC004) and permeabilized with Flow Cytometry Permeabilization/Wash Buffer I (Catalog # FC005). View our protocol for Staining Intracellular Molecules.

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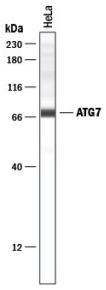
Human/Mouse ATG7 Antibody

Monoclonal Mouse IgG_1 Clone # 683906

Catalog Number: MAB6608

Simple Western



Detection of Human ATG7 by Simple Western™. Simple Western lane view shows lysates of HeLa human cervical epithelial carcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for ATG7 at approximately 72 kDa (as indicated) using 20 µg/mL of Mouse Anti-Human/Mouse ATG7 Monoclonal Antibody (Catalog # MAB6608). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

PREPARATION AND STORAGE

Reconstitution	Sterile PBS to a final concentration of 0.5 mg/mL.
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

ATG7, also known as APG7, is a 78 kDa cytosolic ubiquitin-E1-like enzyme that plays a key role in the autophagic pathway of intracellular bulk degradation. It is required for the conjugation of ATG5 to ATG12, the lipidation of ATG8, and subsequent autophagosome formation. ATG7 is required for mitochondrial removal during erythropoiesis and for the maintenance of axonal homeostasis. Alternate splicing of human ATG7 generates an isoform that lacks 31 aa at the C-terminus, a region that is required for ATG8 lipidation. Human ATG7 shares 93% aa sequence identitiy with mouse and rat ATG7.

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