

Anti-ATG12 Antibody [JE30-61]

HA721504



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, FC
Molecular Wt:	Predicted band size: 15 kDa
Clone number:	JE30-61

Description: Autophagy related 12 is a protein that in humans is encoded by the ATG12 gene. Autophagy is a process of bulk protein degradation in which cytoplasmic components, including organelles, are enclosed in double-membrane structures called autophagosomes and delivered to lysosomes or vacuoles for degradation. ATG12 is the human homolog of a yeast protein involved in autophagy (Mizushima et al., 1998). Autophagy requires the covalent attachment of the protein Atg12 to ATG5 through a ubiquitin-like conjugation system. The Atg12-Atg5 conjugate then promotes the conjugation of ATG8 to the lipid phosphatidylethanolamine. Atg12 was found to be involved in apoptosis. This protein promotes apoptosis through an interaction with anti-apoptotic members of the Bcl-2 family.

Immunogen: Synthetic peptide.

Positive control: HeLa cell lysate, HCT 116 cell lysate, HEK-293 cell lysate, SH-SY5Y cell lysate, HCT 116.

Subcellular location: Cytoplasm, Preautophagosomal structure membrane.

Database links: SwissProt: O94817 Human

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:250
FC	1:500-1:1,000

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4°C after thawing. Aliquot store at -20°C. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

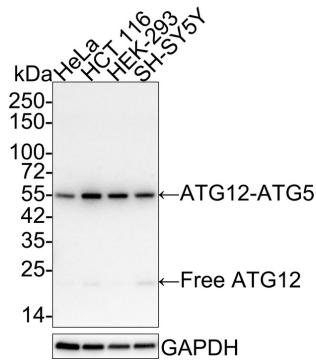
Technical:0086-571-89986345

Service mail:support@huabio.cn

 华安生物
HUABIO
www.huabio.cn

Images

Fig1: Western blot analysis of ATG12 on different lysates with Rabbit anti-ATG12 antibody (HA721504) at 1/1,000 dilution.



Lane 1: HeLa cell lysate
Lane 2: HCT 116 cell lysate
Lane 3: HEK-293 cell lysate
Lane 4: SH-SY5Y cell lysate

Lysates/proteins at 20 µg/Lane.

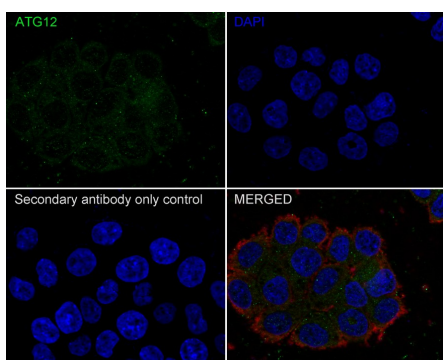
Predicted band size: 15 kDa
Observed band size: 55/20 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDN/TBST for 1 hour at room temperature. The primary antibody (HA721504) at 1/1,000 dilution was used in 5% NFDN/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:100,000 dilution was used for 1 hour at room temperature.

Fig2: Immunocytochemistry analysis of HCT 116 cells labeling ATG12 with Rabbit anti-ATG12 antibody (HA721504) at 1/250 dilution.



Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-ATG12 antibody (HA721504) at 1/250 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn

华安生物
HUABIO
www.huabio.cn

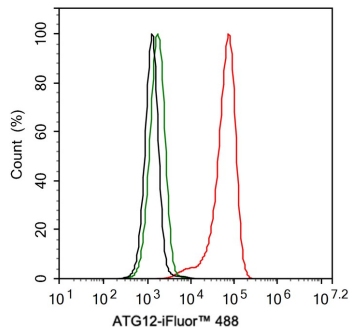


Fig3: Flow cytometric analysis of HCT 116 cells labeling ATG12.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA721504, 1ug/ml) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4 °C for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4 °C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

Background References

1. Lin TY et al. BIRC5/Survivin is a novel ATG12-ATG5 conjugate interactor and an autophagy-induced DNA damage suppressor in human cancer and mouse embryonic fibroblast cells. *Autophagy*. 2020 Jul
2. Keulers TG et al. ATG12 deficiency results in intracellular glutamine depletion, abrogation of tumor hypoxia and a favorable prognosis in cancer. *Autophagy*. 2022 Aug

Hangzhou Huan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn

华安生物
HUABIO
www.huabio.cn