

## Anti-ATG7 Antibody Picoband™

Catalog Number: A00346-5

### About ATG7

Autophagy related 7 is a protein in humans encoded by ATG7 gene. This gene encodes an E1-like activating enzyme that is essential for autophagy and cytoplasmic to vacuole transport. The encoded protein is also thought to modulate p53-dependent cell cycle pathways during prolonged metabolic stress. It has been associated with multiple functions, including axon membrane trafficking, axonal homeostasis, mitophagy, adipose differentiation, and hematopoietic stem cell maintenance. Alternative splicing results in multiple transcript variants.

### Overview

Product Name	Anti-ATG7 Antibody Picoband™
Reactive Species	Human
Description	Boster Bio Anti-ATG7 Antibody Picoband™ catalog # A00346-5. Tested in ELISA, IF, IHC, ICC, WB, Flow Cytometry applications. This antibody reacts with Human.
Application	ELISA, Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal 1B9
Formulation	Each vial contains 4 mg Trehalose, 0.9 mg NaCl, 0.2 mg Na <sub>2</sub> HPO <sub>4</sub> .
Storage Instructions	At -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freezing and thawing.
Host	Rabbit
Uniprot ID	O95352

### Technical Details

Immunogen	E.coli-derived human ATG7 recombinant protein (Position: D71-K645). Human ATG7 shares 92.9% amino acid (aa) sequence identity with both mouse and rat ATG7.
Predicted Reactive Species	Bovine, Canine, Chicken, Primate, Sheep, Xenopus, Zebrafish
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross reactivity with other proteins.
Isotype	IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.

Purification	Immunogen affinity purified.
Suggested Dilutions	<p>Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.</p> <p>If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.</p> <p>Some PubMed article(s) citing the expression level of this target are as follows:</p> <p>Boster Bio's internal QC testing used:</p> <p>Western blot, 0.25-0.5 µg/ml, Human</p> <p>Immunohistochemistry, 2-5 µg/ml, Human</p> <p>Immunocytochemistry/Immunofluorescence, 5 µg/ml, Human</p> <p>Flow Cytometry (Fixed), 1-3 µg/1x10<sup>6</sup> cells, Human</p> <p>ELISA, 0.1-0.5 µg/ml, Human</p>

## Anti-ATG7 Antibody Picoband™ (A00346-5) Images

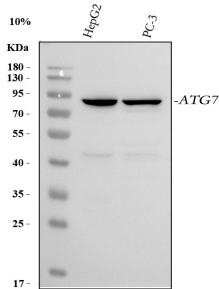


Figure 1. Western blot analysis of ATG7 using anti-ATG7 antibody (A00346-5).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human HepG2 whole cell lysates,

Lane 2: human PC-3 whole cell lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-ATG7 antigen affinity purified polyclonal antibody (Catalog # A00346-5) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for ATG7 at approximately 78 kDa. The expected band size for ATG7 is at 78 kDa.

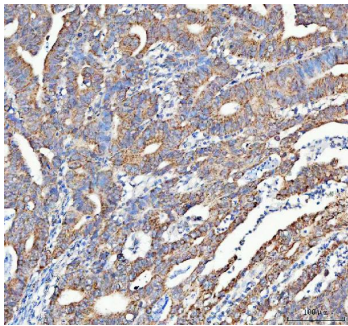


Figure 2. IHC analysis of ATG7 using anti-ATG7 antibody (A00346-5).

ATG7 was detected in a paraffin-embedded section of human colorectal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ATG7 Antibody (A00346-5) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

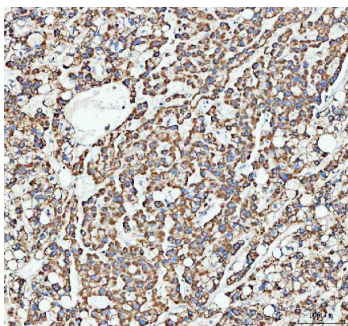
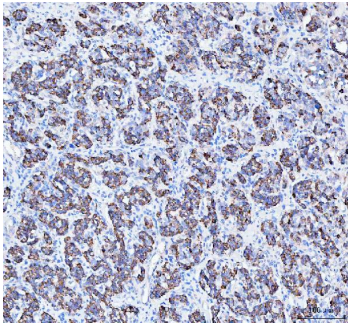


Figure 3. IHC analysis of ATG7 using anti-ATG7 antibody (A00346-5).

ATG7 was detected in a paraffin-embedded section of human liver cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ATG7 Antibody (A00346-5) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

Figure 4. IHC analysis of ATG7 using anti-ATG7 antibody (A00346-5).



ATG7 was detected in a paraffin-embedded section of human lung adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ATG7 Antibody (A00346-5) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

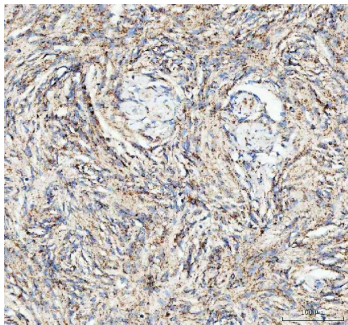


Figure 5. IHC analysis of ATG7 using anti-ATG7 antibody (A00346-5).

ATG7 was detected in a paraffin-embedded section of human meningioma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ATG7 Antibody (A00346-5) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

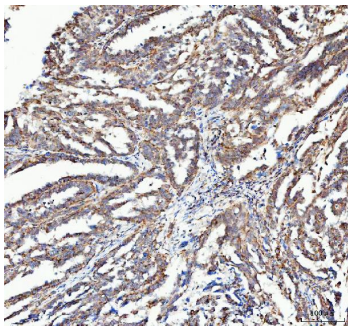


Figure 6. IHC analysis of ATG7 using anti-ATG7 antibody (A00346-5).

ATG7 was detected in a paraffin-embedded section of human ovarian serous adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ATG7 Antibody (A00346-5) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

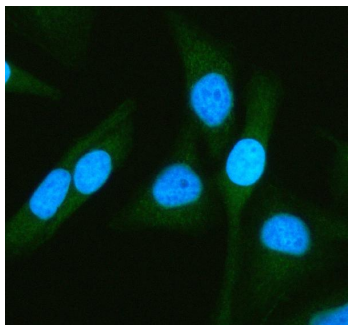
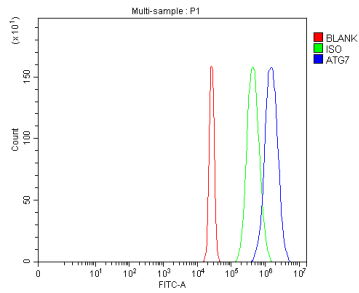


Figure 7. IF analysis of ATG7 using anti-ATG7 antibody (A00346-5).

ATG7 was detected in an immunocytochemical section of HELA cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 ug/mL rabbit anti-ATG7 Antibody (A00346-5) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

Figure 8. Flow Cytometry analysis of HepG2 cells using anti-ATG7 antibody (A00346-5).



Overlay histogram showing HepG2 cells stained with A00346-5 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ATG7 Antibody (A00346-5, 1 ug/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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