

Anti-ATG4A Antibody Picoband™

Catalog Number: A06539-2

About ATG4A

Cysteine protease ATG4A is an enzyme that in humans is encoded by the ATG4A gene. It is mapped to Xq22.3. Autophagy is the process by which endogenous proteins and damaged organelles are destroyed intracellularly. Autophagy is postulated to be essential for cell homeostasis and cell remodeling during differentiation, metamorphosis, non-apoptotic cell death, and aging. Reduced levels of autophagy have been described in some malignant tumors, and a role for autophagy in controlling the unregulated cell growth linked to cancer has been proposed. This gene encodes a member of the autophagin protein family. The encoded protein is also designated as a member of the C-54 family of cysteine proteases.

Overview

Product Name	Anti-ATG4A Antibody Picoband™
Reactive Species	Human
Description	Boster Bio Anti-ATG4A Antibody Picoband™ catalog # A06539-2. Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human.
Application	Flow Cytometry, IHC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Storage Instructions	Store 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 freeze-thaw cycles.
Host	Rabbit
Uniprot ID	Q8WYN0

Technical Details

Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human ATG4A, which shares 75% amino acid (aa) sequence identity with both mouse and rat ATG4A.
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).
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.



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Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit. If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples. Some PubMed article(s) citing the expression level of this target are as follows: Boster Bio's internal QC testing used: Western blot, 0.25-0.5ug/ml, Human Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human Flow Cytometry, 1-3ug/1x10 ⁶ cells, Human
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Anti-ATG4A Antibody Picoband™ (A06539-2) Images

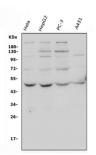


Figure 1. Western blot analysis of ATG4A using anti-ATG4A antibody (A06539-2).

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 50ug of sample under reducing conditions.

Lane 1: human Hela whole cell lysates,

Lane 2: human HepG2 whole cell lysates,

Lane 3: human PC-3 whole cell lysates,

Lane 4: human A431 whole cell lysates.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA 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-ATG4A antigen affinity purified polyclonal antibody (Catalog # A06539-2) 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 1:10000 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 ATG4A at approximately 50KD. The expected band size for ATG4A is at 45KD.

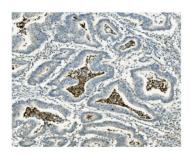


Figure 2. IHC analysis of ATG4A using anti-ATG4A antibody (A06539-2).

ATG4A was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-ATG4A Antibody (A06539-2) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

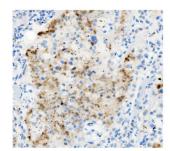
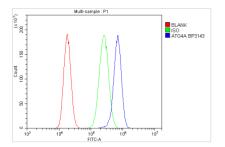


Figure 3. IHC analysis of ATG4A using anti-ATG4A antibody (A06539-2).

ATG4A was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-ATG4A Antibody (A06539-2) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

Figure 4. Flow Cytometry analysis of PC-3 cells using anti-ATG4A antibody (A06539-2).





Overlay histogram showing PC-3 cells stained with A06539-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ATG4A Antibody (A06539-2,1ug/1x10 6 cells) for 30 min at 20 $^\circ$ C. DyLight® 488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10 6 cells) was used as secondary antibody for 30 minutes at 20 $^\circ$ C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x10 6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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Anti-ATG4A Antibody ™