BOSTER
antibody and ELISA experts

BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

# Anti-ATG14L Antibody Picoband™

**Catalog Number:** PB9481

## About ATG14

ATG14 (also known as beclin-1-associated autophagy-related key regulator (Barkor) or ATG14L), an essential autophagy-specific regulator of the class III phosphatidylinositol 3-kinase complex, promotes membrane tethering of protein-free liposomes, and enhances hemifusion and full fusion of proteoliposomes reconstituted with the target (t)-SNAREs (soluble N-ethylmaleimide-sensitive factor attachment protein receptors) syntaxin 17 (STX17) and SNAP29, and the vesicle (v)-SNARE VAMP8 (vesicle-associated membrane protein 8). ATG14 binds to the SNARE core domain of STX17 through its coiled-coil domain, and stabilizes the STX17-SNAP29 binary t-SNARE complex on autophagosomes.

## Overview

| | |
|---|---|
| Product Name | Anti-ATG14L Antibody Picoband™ |
| Reactive Species | Human, Rat |
| Description | Boster Bio Anti-ATG14L Antibody Picoband™ catalog # PB9481. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Rat. |
| Application | Flow Cytometry, IF, IHC, ICC, WB |
| Clonality | Polyclonal |
| Formulation | Each vial contains 5mg BSA, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3. |
| 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 | Q6ZNE5 |

## Technical Details

| | |
|---|---|
| Immunogen | A synthetic peptide corresponding to a sequence at the N-terminus of human ATG14L, different from the related mouse and rat sequences by two amino acids. |
| Predicted Reactive Species | Hamster |
| 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), IHC(F) and ICC. |
| Cross Reactivity | No cross-reactivity with other proteins |
| Isotype | Rabbit IgG |
| Form | Lyophilized |

BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

| | |
|---|---|
| Concentration | Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml. |
| Purification | Immunogen affinity purified. |
| Suggested Dilutions | Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.<br>If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.<br>Some PubMed article(s) citing the expression level of this target are as follows:<br>Boster Bio's internal QC testing used:<br>Western blot, 0.1-0.5ug/ml, Human, Rat<br>Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human, Rat, By Heat<br>Immunocytochemistry/Immunofluorescence, 5 ug/ml, Human<br>Immunofluorescence, 5 ug/ml, Human, Rat<br>Flow Cytometry, 1-3ug/1x$10^6$ cells, Human |

BOSTER
antibody and ELISA experts

BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com

# Anti-ATG14L Antibody Picoband™ (PB9481) Images


Figure 1. Western blot analysis of ATG14L using anti-ATG14L antibody (PB9481).
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: Rat Brain Tissue Lysate,
Lane 2: HELA Whole Cell Lysate.
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-ATG14L antigen affinity purified polyclonal antibody (Catalog # PB9481) 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 ATG14L at approximately 59KD. The expected band size for ATG14L is at 59KD.

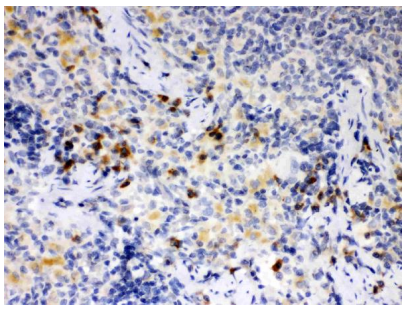


Figure 2. IHC analysis of ATG14L using anti-ATG14L antibody (PB9481).
ATG14L was detected in paraffin-embedded section of Rat Spleen Tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-ATG14L Antibody (PB9481) 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.

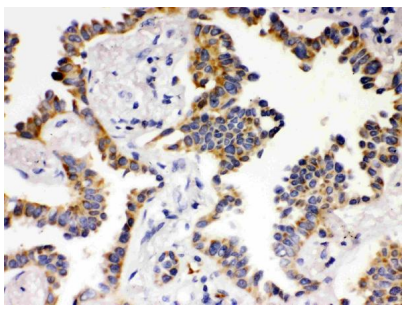
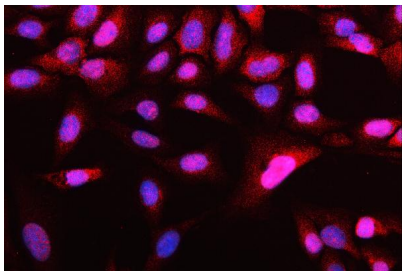


Figure 3. IHC analysis of ATG14L using anti-ATG14L antibody (PB9481).
ATG14L was detected in paraffin-embedded section of Human Lung Cancer Tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-ATG14L Antibody (PB9481) 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. IF analysis of ATG14L using anti-ATG14L antibody (PB9481).
ATG14L was detected in an immunocytochemical section of U2OS cells. Enzyme antigen retrieval was performed using

BOSTER BIOLOGICAL TECHNOLOGY
3942 B Valley Ave, Pleasanton, CA 94566
888-466-3604 | support@bosterbio.com | www.bosterbio.com



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-ATG14L Antibody (PB9481) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) 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.

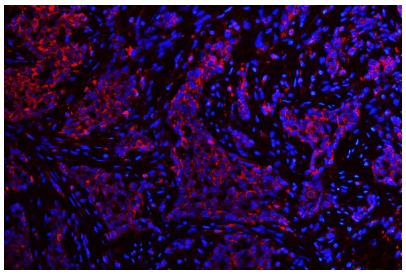


Figure 5. IF analysis of ATG14L using anti-ATG14L antibody (PB9481).
ATG14L was detected in a paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 ug/mL rabbit anti-ATG14L Antibody (PB9481) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) 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.

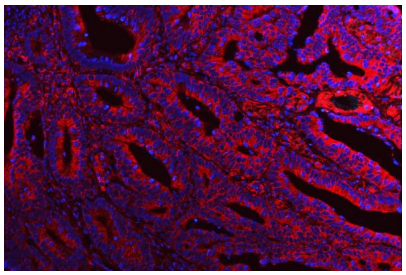


Figure 6. IF analysis of ATG14L using anti-ATG14L antibody (PB9481).
ATG14L was detected in a paraffin-embedded section of human colon cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 ug/mL rabbit anti-ATG14L Antibody (PB9481) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) 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.

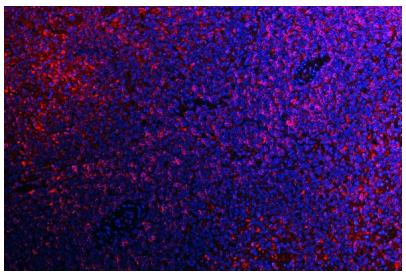
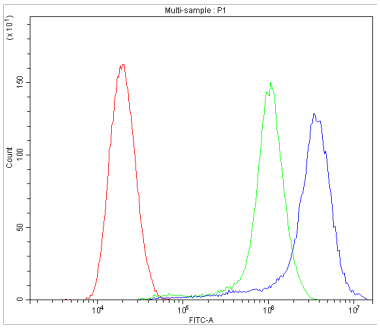


Figure 7. IF analysis of ATG14L using anti-ATG14L antibody (PB9481).
ATG14L was detected in a paraffin-embedded section of rat spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 ug/mL rabbit anti-ATG14L Antibody (PB9481) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) 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 SiHa cells using anti-ATG14L antibody (PB9481).
Overlay histogram showing SiHa cells stained with PB9481 (Blue line).The cells were blocked with 10% normal goat




serum. And then incubated with rabbit anti-ATG14L Antibody (PB9481,1ug/1x10$^6$ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10$^6$ cells) was used as secondary antibody for 30 minutes at 20°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.

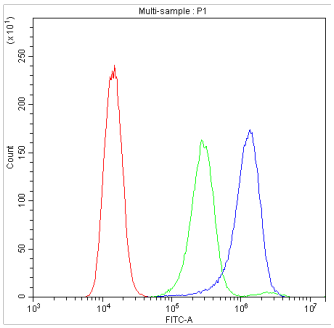



Figure 9. Flow Cytometry analysis of A431 cells using anti-ATG14L antibody (PB9481).
Overlay histogram showing A431 cells stained with PB9481 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ATG14L Antibody (PB9481,1ug/1x10$^6$ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10$^6$ cells) was used as secondary antibody for 30 minutes at 20°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.

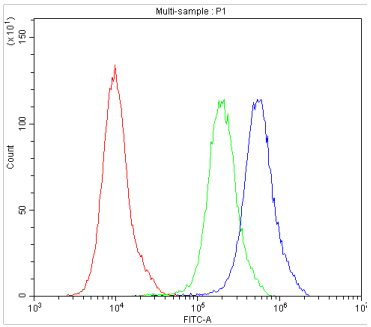



Figure 10. Flow Cytometry analysis of PC-3 cells using anti-ATG14L antibody (PB9481).
Overlay histogram showing PC-3 cells stained with PB9481 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ATG14L Antibody (PB9481,1ug/1x10$^6$ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10$^6$ cells) was used as secondary antibody for 30 minutes at 20°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.

## 1 Publications Citing This Product

1. PubMed ID: 10.1007/s11418-020-01399-5, Polydatin induces apoptosis and autophagy via STAT3 signaling in human osteosarcoma MG-63 cells

Visit bosterbio.com/anti-atg14l-picoband-trade-antibody-pb9481-boster.html to see all 1 publications.