

## Anti-FOXO3A Antibody Picoband®

Catalog Number: PB9196

### About FOXO3

Forkhead box O3, also known as FKHRL1 or FOXO3a, is a human protein encoded by the FOXO3 gene. FOXO3 belongs to the O subclass of the forkhead family of transcription factors which are characterized by a distinct fork head DNA-binding domain. It is mapped to 6q21. This protein likely functions as a trigger for apoptosis through upregulation of genes necessary for cell death, such as Bim and PUMA, or downregulation of anti-apoptotic proteins such as FLIP. In mammals FOXO3 regulates the resistance of cells to stress by inducing DNA repair and thereby may also affect organismal life span. In addition, it is thought that FOXO3 is also involved in protection from oxidative stress by upregulating antioxidants such as catalase and MnSOD.

### Overview

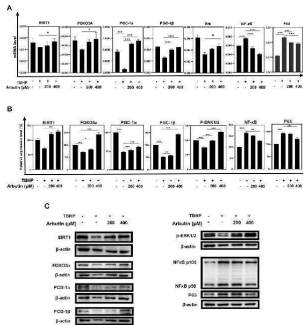
Product Name	Anti-FOXO3A Antibody Picoband®
Reactive Species	Human, Rat
Description	Boster Bio Anti-FOXO3A Antibody Picoband® catalog # PB9196. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Rat. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na <sub>2</sub> HPO <sub>4</sub> .
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	O43524

### Technical Details

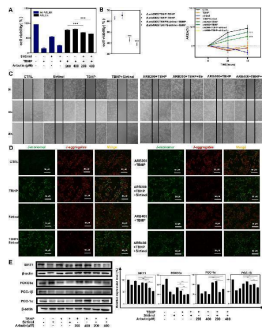
Immunogen	E.coli-derived human FOXO3A recombinant protein (Position: Q471-G673). Human FOXO3A shares 97% amino acid (aa) sequence identity with mouse FOXO3A.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins
Isotype	Rabbit IgG

Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.1-0.5ug/ml, Human, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human Immunocytochemistry/Immunofluorescence, 5 ug/ml, Human Flow Cytometry(Fixed), 1-3 ug/1x10 <sup>6</sup> cells, Human

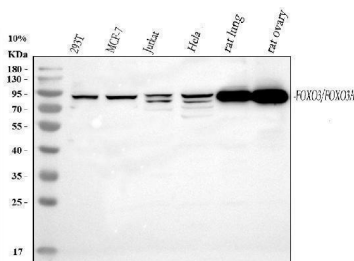
## Anti-FOXO3A Antibody Picoband® (PB9196) Images



Arbutin exerted protective effects via the SIRT1/FOXO3a/PGC-1alpha/beta and NF-kappaB/p65 signaling pathway. (A) qRT-PCR was used to measure transcript levels of the SIRT1/FOXO3a/PGC-1alpha/beta pathway and NF-kappaB/p65 genes. TBHP decreased the expression of SIRT1, FOXO3a, and PGC-1alpha/beta and increased the expression of NF-kappaB/p65, whereas mRNA levels in the groups that were pretreated with Arbutin showed reversed trend. (B) western blots were conducted to detect the proteins level of SIRT1, FOXO3a, PGC-1alpha/beta, p-ERK, and NFKB1/P65. (C) imagej was used to analyze the relative expression level of the proteins mentioned above (\* p < 0.05, \*\* p < 0.01, \*\*\* p < 0.001, n = 3, bars represent SD). Index in PubMed under a CC BY license. PMID: 36051689

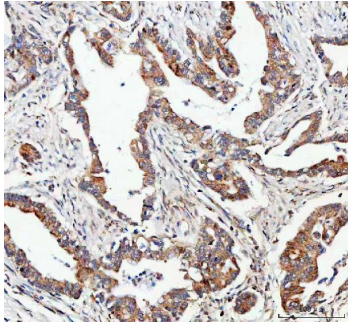


Sirtinol diminished the capability of Arbutin to assist ARPE-19 cells to defend against oxidative stress. (A) (B) flow cytometric analysis showed that cells treated with only sirtinol, TBHP, cotreated with sirtinol, and TBHP displayed decreased cellular viability. However, sirtinol conduction diminished the protective capacity of Arbutin. (C) ARPE-19 cells were seeded in a 24-well plate and applied wounds at the confluence of 80%. The cells were pretreated with or without Arbutin and then subjected to TBHP (350 μM); meanwhile, cells in certain groups were incubated with sirtinol. Photos were taken at different time points post distinct treatments. (D) fluorescence images observed that ARPE-19 treated with Arbutin while subjected to sirtinol and then exposed to TBHP was unable to recuperate ΔΨm. (E) with sirtinol administration, the protein levels of the SIRT1/FOXO3a/PGC-1alpha/beta pathway decreased in the presence of Arbutin (\* p < 0.05, \*\* p < 0.01, \*\*\* p < 0.001, n = 3, bars represent SD). Index in PubMed under a CC BY license. PMID: 36051689

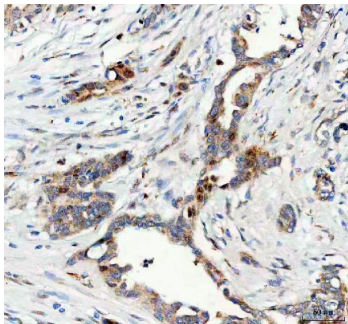


Western blot analysis of FOXO3A using anti-FOXO3A antibody (PB9196). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human 293T whole cell lysates, Lane 2: human MCF-7 whole cell lysates, Lane 3: human Jurkat whole cell lysates, Lane 4: human Hela whole cell lysates, Lane 5: rat lung tissue lysates, Lane 6: rat ovary tissue 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-FOXO3A antigen affinity purified polyclonal antibody (Catalog # PB9196) at 0.5 ug/mL overnight at 4°C, then washed with 0.5 TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a

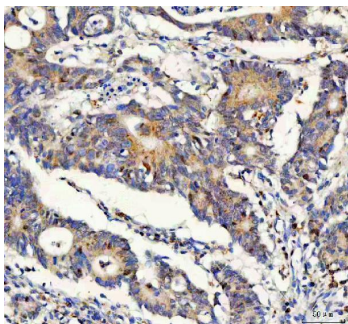
dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for FOXO3A at approximately 80-90 kDa. The expected band size for FOXO3A is at 71 kDa.



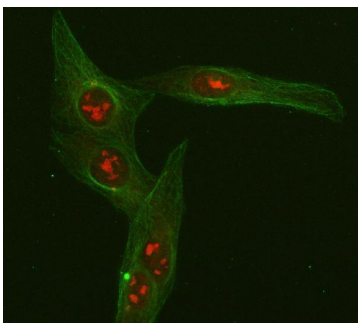
IHC analysis of FOXO3A using anti-FOXO3A antibody (PB9196). FOXO3A was detected in a paraffin-embedded section of human pancreatic ductal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-FOXO3A Antibody (PB9196) 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.



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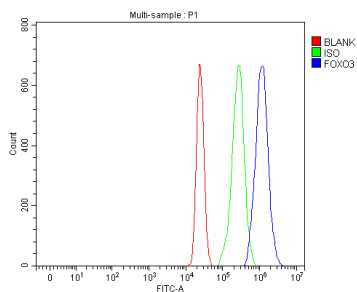


IHC analysis of FOXO3A using anti-FOXO3A antibody (PB9196). FOXO3A was detected in a paraffin-embedded section of human colorectal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-FOXO3A Antibody (PB9196) 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.



IF analysis of FOXO3A using anti-FOXO3A antibody (PB9196) and anti-Beta Tubulin antibody (M01857-3). FOXO3A was detected in immunocytochemical section of U2OS cell. 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-FOXO3A Antibody (PB9196) and mouse anti-Beta Tubulin antibody (M01857-3) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) and DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) were used as secondary antibody at 1:500 dilution and incubated for 30

minutes at 37°C. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



Flow Cytometry analysis of MCF-7 cells using anti-FOXO3A antibody (PB9196). Overlay histogram showing MCF-7 cells stained with PB9196 (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-FOXO3A Antibody (PB9196, 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.

## 1 Publications Citing This Product

1. PubMed ID: 10.3390/ijms22042225, Regulation of Glucose Metabolism by MuRF1 and Treatment of Myopathy in Diabetic Mice with Small Molecules Targeting MuRF1

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