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Anti-ULK3 Antibody Picoband®

Catalog Number: PB9519

About ULK3

Serine/threonine protein kinase that acts as a regulator of Sonic hedgehog (SHH) signaling and autophagy. In search of potential homologues to Drosophila and human Fu, people have cloned human serine/threonine kinase ULK3 and assessed its ability to regulate GLI transcription factors, mediators of SHH signaling. And ULK3 enhances endogenous and over-expressed GLI1 and GLI2 transcriptional activity in cultured cells, as assessed by GLI-luciferase reporter assay. Besides that, ULK3 alters subcellular localization of GLI1, as assessed by immunofluorescent staining and immunoblotting assays. It is showed that ULK3 is an autophosphorylated kinase and phosphorylates GLI proteins in vitro. Also, ULK3 catalytical activity is crucial for its function in SHH pathway. It is demonstrated that ULK3 is widely expressed and its expression is higher in a number of tissues where Shh signaling is known to be active. The data suggest that serine/threonine kinase ULK3 is involved in the SHH pathway as a positive regulator of GLI proteins.

Overview

Product Name	Anti-ULK3 Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-ULK3 Antibody Picoband® catalog # PB9519. Tested in Flow Cytometry, IHC, IHC-F, ICC, WB applications. This antibody reacts with Human, Mouse, 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, IHC, IHC-F, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains antibody formulated with stabilizing components, 0.9 mg NaCl, 0.2 mg Na2HPO4, and 0.05 mg NaN3. *This antibody is supplied in a stabilized formulation. Compatibility with conjugation reactions depends on the chemistry of the conjugation method used. For conjugation methods that are not compatible with the stabilizing components present in this formulation, a carrier-free antibody format is required.
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	Q6PHR2

Technical Details

Immunogen	A synthetic peptide corresponding to a sequence at C-terminus of human ULK3, different from the related mouse and rat sequences by four amino acids.

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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.
Suggested Dilutions	Western blot, 0.1-0.5ug/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human, Rat Immunohistochemistry (Frozen Section), 0.5-1ug/ml, Human Immunocytochemistry, 0.5-1ug/ml, Human Flow Cytometry (Fixed), 1-3ug/1x10^6 cells, Human

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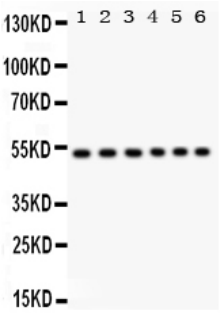
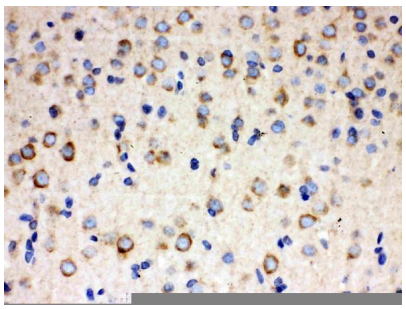
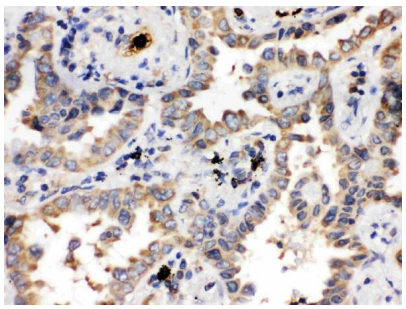
Anti-ULK3 Antibody Picoband® (PB9519) Images



Western blot analysis of ULK3 using anti-ULK3 antibody (PB9519). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. Lane 1: Rat Brain Tissue Lysate at 50ug, Lane 2: Rat Testis Tissue Lysate at 50ug, Lane 3: Mouse Brain Tissue Lysate at 50ug, Lane 4: Human Placenta Tissue Lysate at 50ug, Lane 5: 22RV1 Whole Cell Lysate at 40ug, Lane 6: SMMC Whole Cell Lysate at 40ug. 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-ULK3 antigen affinity purified polyclonal antibody (Catalog # PB9519) 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 ULK3 at approximately 53 kDa. The expected band size for ULK3 is at 53 kDa.



IHC analysis of ULK3 using anti-ULK3 antibody (PB9519). ULK3 was detected in a paraffin-embedded section of rat brain 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 1 ug/ml rabbit anti-ULK3 Antibody (PB9519) 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.



IHC analysis of ULK3 using anti-ULK3 antibody (PB9519). ULK3 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 1 ug/ml rabbit anti-ULK3 Antibody (PB9519) 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.

Flow Cytometry analysis of U20S cells using anti-ULK3 antibody (PB9519). Overlay histogram showing U20S cells stained with PB9519 (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

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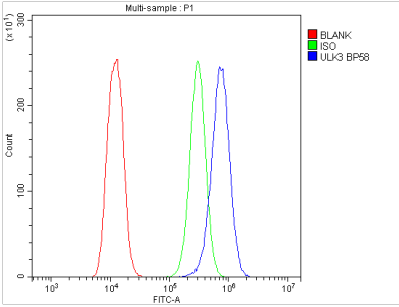
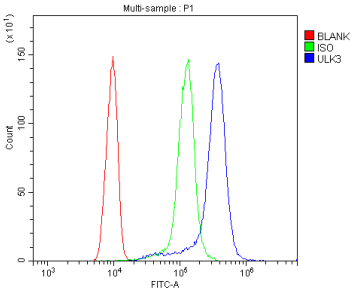


with rabbit anti-ULK3 Antibody (PB9519,1ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



Flow Cytometry analysis of THP-1 cells using anti-ULK3 antibody (PB9519). Overlay histogram showing THP-1 cells stained with PB9519 (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-ULK3 Antibody (PB9519,1ug/1x106 cells) for 30 min at 20°C. DyLight488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

Anti-ULK3 Antibody

For Research Use Only. Not for use in diagnostic procedures.