

Anti-Optineurin/OPTN Antibody Picoband™

Catalog Number: PB9343

About OPTN

OPTN is also known as NRP, FIP2 or HYPL. This gene encodes the coiled-coil containing protein optineurin. Optineurin may play a role in normal-tension glaucoma and adult-onset primary open angle glaucoma. Optineurin interacts with adenovirus E3-14.7K protein and may utilize tumor necrosis factor-alpha or Fas-ligand pathways to mediate apoptosis, inflammation or vasoconstriction. Optineurin may also function in cellular morphogenesis and membrane trafficking, vesicle trafficking, and transcription activation through its interactions with the RAB8, huntingtin, and transcription factor IIIA proteins. Alternative splicing results in multiple transcript variants encoding the same protein.

Overview

Product Name	Anti-Optineurin/OPTN Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-Optineurin/OPTN Antibody Picoband™ catalog # PB9343. Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	Flow Cytometry, IHC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na2HPO4.
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	Q96CV9

Technical Details

Immunogen	E.coli-derived human Optineurin recombinant protein (Position: R241-I577). Human Optineurin shares 82% amino acid (aa) sequence identity with both mouse and rat Optineurin.
Predicted Reactive Species	Bovine
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	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, 0.25-0.5 ug/ml, Human, Mouse, Rat Immunohistochemistry(Paraffin-embedded Section), 2-5 ug/ml, Human, Mouse, Rat Flow Cytometry, 1-3 ug/1x10⁶ cells, Human

Anti-Optineurin/OPTN Antibody Picoband™ (PB9343) Images

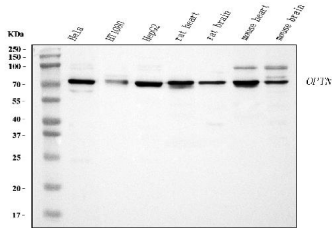


Figure 1. Western blot analysis of Optineurin using anti-Optineurin antibody (PB9343).

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 HeLa whole cell lysates,
Lane 2: human HT1080 whole cell lysates,
Lane 3: human HepG2 whole cell lysates,
Lane 4: rat heart tissue lysates,
Lane 5: rat brain tissue lysates,
Lane 6: mouse heart tissue lysates,
Lane 7: mouse brain 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-Optineurin antigen affinity purified polyclonal antibody (Catalog # PB9343) 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 Optineurin at approximately 75 kDa. The expected band size for Optineurin is at 66 kDa.

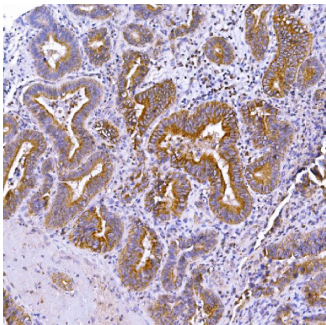


Figure 2. IHC analysis of Optineurin using anti-Optineurin antibody (PB9343).

Optineurin was detected in a paraffin-embedded section of human gall bladder 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-Optineurin Antibody (PB9343) 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 Streptavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

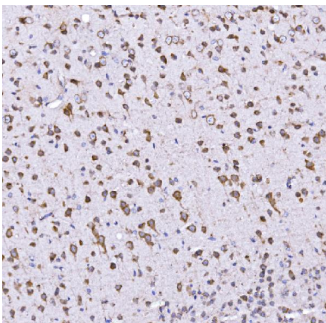


Figure 3. IHC analysis of Optineurin using anti-Optineurin antibody (PB9343).

Optineurin was detected in a paraffin-embedded section of mouse 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 2 ug/ml rabbit anti-Optineurin Antibody (PB9343) 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 Streptavidin-Biotin-Complex

(SABC) (Catalog # SA1022) with DAB as the chromogen.

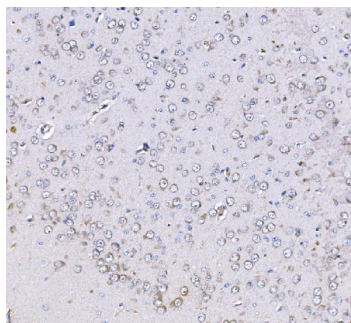


Figure 4. IHC analysis of Optineurin using anti-Optineurin antibody (PB9343).

Optineurin 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 2 ug/ml rabbit anti-Optineurin Antibody (PB9343) 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 Streptavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

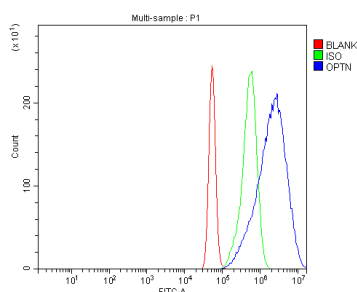


Figure 5. Flow Cytometry analysis of A431 cells using anti-Optineurin antibody (PB9343).

Overlay histogram showing A431 cells stained with PB9343 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-Optineurin Antibody (PB9343, 1 ug/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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