

Anti-CIAS1/NALP3/NLRP3 Antibody

Catalog Number: PA1665

About NLRP3

NLRP3 (NLR FAMILY, PYRIN DOMAIN-CONTAINING 3), also known as CIAS1, CRYOPYRIN, NALP3 or PYPAF1, is a protein that in humans is encoded by the NLRP3 (NOD-like receptor family, pyrin domain containing 3) gene. The NLRP3 gene encodes a pyrin-like protein expressed predominantly in peripheral blood leukocytes. And the NLRP3 gene is mapped on 1q44. NLRP3 interacts with apoptosis-associated speck-like protein containing a CARD (ASC). The encoded protein may play a role in the regulation of inflammation and apoptosis. Mutation of the NALP3 nucleotide-binding domain reduced ATP binding, CASP1 activation, IL1B production, cell death, macromolecular complex formation, self-association, and association with ASC. Consistent with an essential role for Nlrp3 inflammasomes in antifungal immunity, Gross et al. showed that Nlrp3-deficient mice are hypersusceptible to *C. albicans* infection. Activation of the NLRP3 inflammasome in response to virus or to RNA was dependent upon lysosomal maturation and reactive oxygen species production in human cells. The NLRP3 inflammasome senses obesity-associated danger signals and contributes to obesity-induced inflammation and insulin resistance.

Overview

Product Name	Anti-CIAS1/NALP3/NLRP3 Antibody
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-CIAS1/NALP3/NLRP3 Antibody catalog # PA1665. 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 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na ₂ HPO ₄ .
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	Q96P20

Technical Details

Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human CIAS1, different from the related rat and mouse sequences by one amino acid.
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).
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	<p>Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.</p> <p>If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.</p> <p>Some PubMed article(s) citing the expression level of this target are as follows:</p> <p>Boster Bio's internal QC testing used:</p> <p>Western blot, 0.1-0.5ug/ml, Human, Mouse, Rat</p> <p>Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human, By Heat</p> <p>Flow Cytometry(Fixed), 1-3ug/1x10⁶ cells, Human</p>

Anti-CIAS1/NALP3/NLRP3 Antibody (PA1665) Images

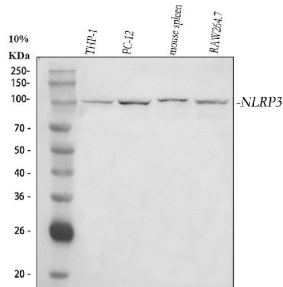


Figure 1. Western blot analysis of NLRP3 using anti-NLRP3 antibody (PA1665).

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 THP-1 whole cell lysates,

Lane 2: rat PC-12 whole cell lysates,

Lane 3: mouse spleen whole cell lysates,

Lane 4: mouse RAW264.7 whole cell 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-NLRP3 antigen affinity purified polyclonal antibody (Catalog # PA1665) 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 NLRP3 at approximately 110 kDa. The expected band size for NLRP3 is at 118 kDa.

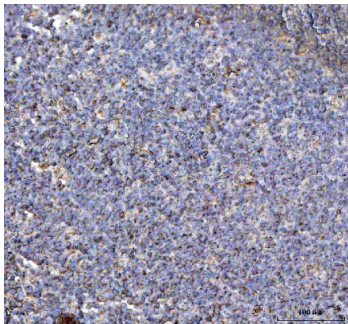


Figure 2. IHC analysis of NLRP3 using anti-NLRP3 antibody (PA1665).

NLRP3 was detected in a paraffin-embedded section of human tonsil 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-NLRP3 Antibody (PA1665) 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.

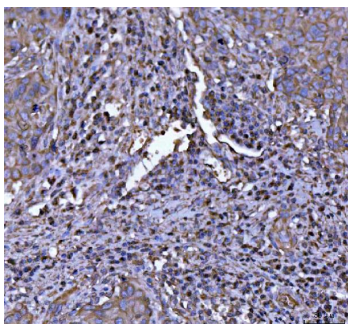


Figure 3. IHC analysis of NLRP3 using anti-NLRP3 antibody (PA1665).

NLRP3 was detected in a paraffin-embedded section of human squamous cell cervical carcinoma 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-NLRP3 Antibody (PA1665) 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.

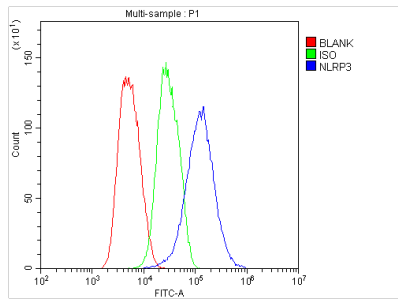


Figure 4. Flow Cytometry analysis of THP-1 cells using anti-NLRP3 antibody (PA1665). Overlay histogram showing THP-1 cells stained with PA1665 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-NLRP3 Antibody (PA1665, 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.

18 Publications Citing This Product

1. PubMed ID: 10.1016/j.cbi.2021.109572, Pelargonic acid vanillylamide and rosuvastatin protect against oxidized low-density lipoprotein-induced endothelial dysfunction by inhibiting the NF-kappaB/NLRP3 pathway and improving cell-cell junctions
2. PubMed ID: 10.1002/jbt.22978, The combination of dapagliflozin and statins ameliorates renal injury through attenuating the activation of inflammasome-mediated autophagy in insulin-resistant rats
3. PubMed ID: 34217687, Sivasinprasasn S,Wikan N,Tocharus J,Chaichompoo W,Suksamrarn A,Tocharus C.Pelargonic acid vanillylamide and rosuvastatin protect against oxidized low-density lipoprotein-induced endothelial dysfunction by inhibiting the NF-kappaB/NLRP3 pathway and improving cell-cell junctions.Chem Biol Interact.2021 Jul 1:109572.doi:10.1016/j.cbi.2021.109572.Epub ahead of print.PMID:34217687.

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