

## Anti-HSPA4 Antibody Picoband®

Catalog Number: A03618-2

### About HSPA4

Heat shock 70 kDa protein 4 is a protein that in humans is encoded by the HSPA4 gene. The heat shock protein HSPA4 (Apg-2, HSP70RY) is a member of the heat-shock protein 110 (Hsp 110) subfamily of Hsp70 heat-shock proteins. Apg-2 has chaperone ability similar to Hsp110, and it plays a role under non-stress conditions. Apg-2 interacts with TJP1/ZO-1 and functions as a regulator of ZO-1-ZONAB signaling in epithelial cells in response to cellular stress.

### Overview

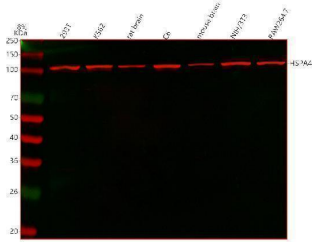
Product Name	Anti-HSPA4 Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-HSPA4 Antibody Picoband® catalog # A03618-2. Tested in WB, IHC, ICC/IF, FCM applications. This antibody reacts with Human, Mouse, Rat. The brand Picoband indicates this is a premium antibody that 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, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4 mg Trehalose, 0.9 mg NaCl, 0.2 mg Na <sub>2</sub> HPO <sub>4</sub> .
Storage Instructions	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 freezing and thawing.
Host	Rabbit
Uniprot ID	P34932

### Technical Details

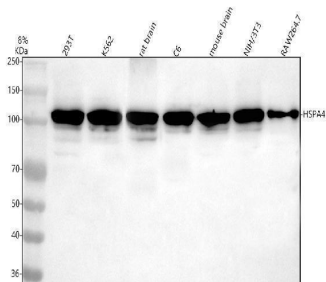
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human HSPA4, which shares 95.2% amino acid (aa) sequence identity with both mouse and rat HSPA4.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
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, 0.1-0.25 ug/ml, Human, Mouse, Rat

Immunohistochemistry(Paraffin-embedded Section), 2-5 ug/ml, Human, Mouse  
Immunocytochemistry/Immunofluorescence, 5 ug/ml, Human  
Flow Cytometry (Fixed), 1-3 ug/1x<sup>6</sup> cells, Human

## Anti-HSPA4 Antibody Picoband® (A03618-2) Images

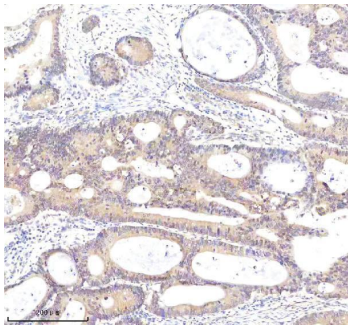


Western blot analysis of HSPA4 using anti-HSPA4 antibody (A03618-2). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human 293T whole cell lysates, Lane 2: human K562 whole cell lysates, Lane 3: rat brain tissue lysates, Lane 4: rat C6 whole cell lysates, Lane 6: mouse brain tissue lysates, Lane 7: mouse NIH/3T3 whole cell lysates, Lane 8: 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-HSPA4 antigen affinity purified polyclonal antibody (Catalog # A03618-2) at 0.25 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-DyLight 647 Conjugated secondary antibody at a dilution of 1:2000 for 1.5 hour at RT. A specific band was detected for HSPA4 at approximately 110 kDa. The expected band size for HSPA4 is at 96,14 kDa.

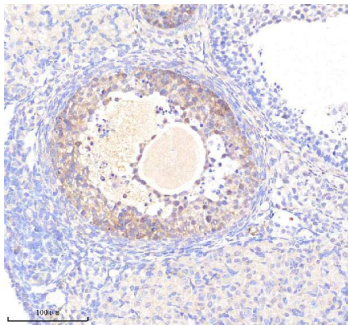


Western blot analysis of HSPA4 using anti-HSPA4 antibody (A03618-2). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human 293T whole cell lysates, Lane 2: human K562 whole cell lysates, Lane 3: rat brain tissue lysates, Lane 4: rat C6 whole cell lysates, Lane 5: mouse brain tissue lysates, Lane 6: mouse NIH/3T3 whole cell lysates, Lane 7: 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-HSPA4 antigen affinity purified polyclonal antibody (Catalog # A03618-2) 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 HSPA4 at approximately 110 kDa. The expected band size for HSPA4 is at 94,16 kDa.

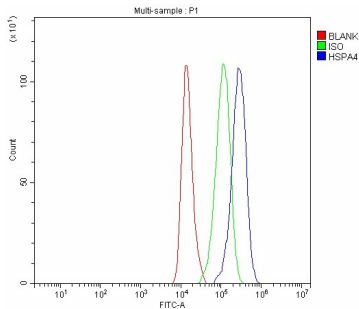
IHC analysis of HSPA4 using anti-HSPA4 antibody (A03618-2). HSPA4 was detected in a paraffin-embedded section of human colon cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-HSPA4 Antibody (A03618-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit



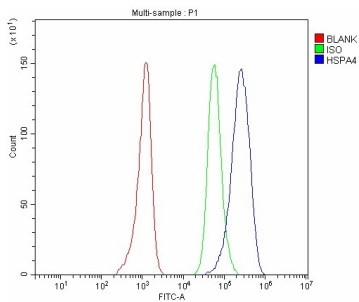
IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



IHC analysis of HSPA4 using anti-HSPA4 antibody (A03618-2). HSPA4 was detected in a paraffin-embedded section of mouse ovary 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-HSPA4 Antibody (A03618-2) 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.



Flow Cytometry analysis of 293T cells using anti-HSPA4 antibody (A03618-2). Overlay histogram showing 293T cells stained with A03618-2 (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-HSPA4 Antibody (A03618-2, 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 without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



Flow Cytometry analysis of MOLT-4 cells using anti-HSPA4 antibody (A03618-2). Overlay histogram showing MOLT-4 cells stained with A03618-2 (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-HSPA4 Antibody (A03618-2, 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 without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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### Anti-HSPA4 Antibody

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