

Anti-CD272/BTLA Antibody Picoband™

Catalog Number: A03149

About BTLA

B- and T-lymphocyte attenuator is a protein that in humans is encoded by the BTLA gene. BTLA has also been designated as CD272 (cluster of differentiation 272). This gene encodes a member of the immunoglobulin superfamily. The encoded protein contains a single immunoglobulin (Ig) domain and is a receptor that relays inhibitory signals to suppress the immune response. Alternative splicing results in multiple transcript variants. Polymorphisms in this gene have been associated with an increased risk of rheumatoid arthritis. BTLA expression is induced during activation of T cells, and BTLA remains expressed on Th1 cells but not Th2 cells. Like PD1 and CTLA4, BTLA interacts with a B7 homolog, B7H4.

Overview

Product Name	Anti-CD272/BTLA Antibody Picoband™
Reactive Species	Human, Mouse
Description	Boster Bio Anti-CD272/BTLA Antibody Picoband™ catalog # A03149. Tested in Flow Cytometry, IHC, IHC-F, ICC, WB applications. This antibody reacts with Human, Mouse.
Application	Flow Cytometry, IHC, IHC-F, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
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	Q7Z6A9

Technical Details

Immunogen	A synthetic peptide corresponding to a sequence in the middle region of human CD272/BTLA.
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), IHC(F) 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

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, 0.1-0.5ug/ml

Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml

Immunohistochemistry (Frozen Section), 0.5-1ug/ml

Immunocytochemistry, 0.5-1ug/ml

Flow Cytometry, 1-3ug/1x10⁶ cells

Anti-CD272/BTLA Antibody Picoband™ (A03149) Images

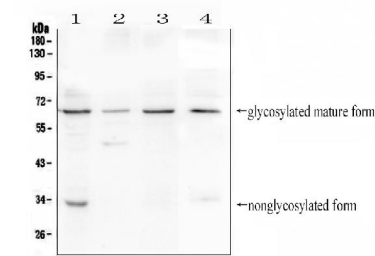


Figure 1. Western blot analysis of CD272 using anti-CD272 antibody (A03149).

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 50ug of sample under reducing conditions.

Lane 1: human HEK293 whole cell lysate,

Lane 2: human Jurkat whole cell lysate,

Lane 3: human CCRF-CEM whole cell lysate,

Lane 4: mouse thymus tissue lysate.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA 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-CD272 antigen affinity purified polyclonal antibody (Catalog # A03149) 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 1:10000 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 CD272 at approximately 65KD. The expected band size for CD272 is at 33KD.

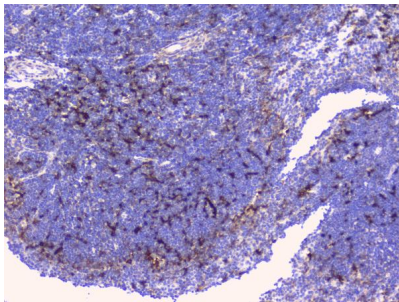


Figure 2. IHC analysis of CD272/BTLA using anti-CD272/BTLA antibody (A03149).

CD272/BTLA was detected in paraffin-embedded section of mouse spleen tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-CD272/BTLA Antibody (A03149) 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.

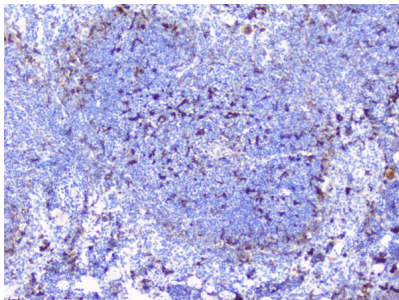
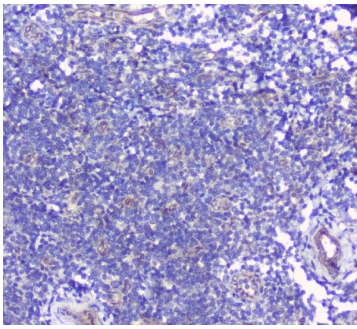


Figure 3. IHC analysis of CD272/BTLA using anti-CD272/BTLA antibody (A03149).

CD272/BTLA was detected in paraffin-embedded section of mouse spleen tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-CD272/BTLA Antibody (A03149) 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 CD272/BTLA using anti-CD272/BTLA antibody (A03149).



CD272/BTLA was detected in paraffin-embedded section of human tonsil tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-CD272/BTLA Antibody (A03149) 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.

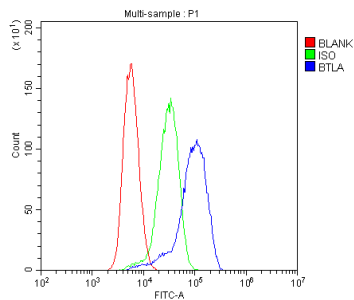


Figure 5. Flow Cytometry analysis of THP-1 cells using anti-CD272/BTLA antibody (A03149).

Overlay histogram showing THP-1 cells stained with A03149 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-CD272/BTLA Antibody (A03149, 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 (Red line) was also used as a control.

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