

Anti-CD9 Antibody Picoband™

Catalog Number: PB9930

About Cd9

CD9 antigen is a protein that in humans is encoded by the CD9 gene. CD9 is a cell surface glycoprotein that is known to complex with integrins and other transmembrane 4 superfamily proteins. It is found on the surface of exosomes. It can modulate cell adhesion and migration and also trigger platelet activation and aggregation. In addition, the protein appears to promote muscle cell fusion and support myotube maintenance. This protein also seems to be a key part in the egg-sperm fusion during mammalian fertilization. While oocytes are ovulated, CD9-deficient oocytes are not properly fused with sperm upon fertilization. CD9 is located in the microvillar membrane of the oocytes and also appears to intervene in maintaining the normal shape of oocyte microvilli.

Overview

Product Name	Anti-CD9 Antibody Picoband™
Reactive Species	Mouse, Rat
Description	Boster Bio Anti-CD9 Antibody Picoband™ catalog # PB9930. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Mouse, Rat.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 5mg BSA, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
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	P40240

Technical Details

Immunogen	E. coli-derived mouse CD9 recombinant protein (Position: T110-I193). Mouse CD9 shares 77.4% and 86.9% amino acid (aa) sequence identity with human and rat CD9, respectively.
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) 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	<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, Mouse, Rat</p> <p>Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Mouse, By Heat</p> <p>Immunocytochemistry/Immunofluorescence, 5ug/ml, Mouse</p> <p>Flow Cytometry, 1-3 ug/1x10⁶ cells, Mouse</p>

Anti-CD9 Antibody Picoband™ (PB9930) Images



Figure 1. Western blot analysis of CD9 using anti-CD9 antibody (PB9930). 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: mouse kidney 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-CD9 antigen affinity purified polyclonal antibody (Catalog # PB9930) 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 CD9 at approximately 25 kDa. The expected band size for CD9 is at 25 kDa.

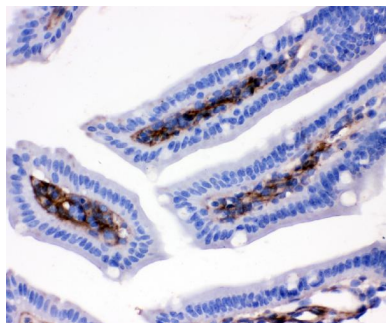


Figure 2. IHC analysis of CD9 using anti-CD9 antibody (PB9930).

CD9 was detected in paraffin-embedded section of mouse intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-CD9 Antibody (PB9930) 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.

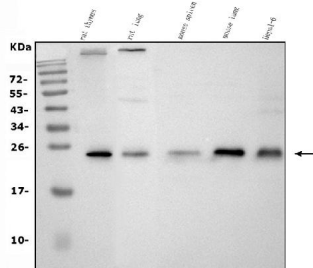


Figure 3. Western blot analysis of CD9 using anti-CD9 antibody (PB9930).

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 30ug of sample under reducing conditions.

Lane 1: rat thymus tissue lysates,

Lane 2: rat lung tissue lysates,

Lane 3: mouse spleen tissue lysates,

Lane 4: mouse lung tissue lysates,

Lane 5: mouse HEPA1-6 whole cell lysates.

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-CD9 antigen affinity purified polyclonal antibody (Catalog # PB9930) 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 CD9 at approximately 25KD. The expected band size for CD9 is at 25KD.

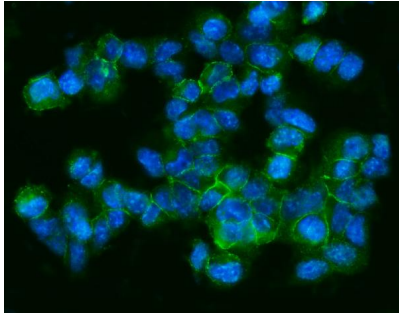


Figure 4. IF analysis of CD9 using anti-CD9 antibody (PB9930).

CD9 was detected in immunocytochemical section of HEPA1-6 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5ug/mL rabbit anti-CD9 Antibody (PB9930) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

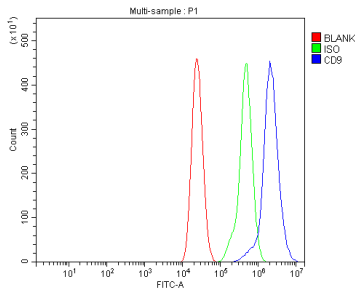


Figure 5. Flow Cytometry analysis of RAW264.7 cells using anti-CD9 antibody (PB9930).

Overlay histogram showing RAW264.7 cells stained with PB9930 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-CD9 Antibody (PB9930, 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.

2 Publications Citing This Product

1. PubMed ID: 32059163, Wang D,Hao C,Zhang L,Zhang J,Liu S,Li Y,Qu Y,Zhao Y,Huang R,Wei J,Yao W.Exosomal miR-125a-5p derived from silica-exposed macrophages induces fibroblast transdifferentiation.Ecotoxicol Environ Saf.2020 Apr 1;192:110253.doi:10.1016/j.ecoenv.2020.110253.Epub

2. PubMed ID: 32400849, Cao G,Meng X, Han X,Li J.Exosomes derived from circRNA Rtn4-modified BMSCs attenuate TNF-alpha-induced cytotoxicity and apoptosis in murine MC3T3-E1 cells by sponging miR-146a.Biosci Rep.2020 May 29;40(5): BSR20193436.doi:10.1042/BSR20193436.PMID:32400849;PMC

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