

Anti-CD146/MCAM Antibody Picoband™ (monoclonal, 4C12)

Catalog Number: M01683-3

About MCAM

CD146 (cluster of differentiation 146), also known as the melanoma cell adhesion molecule (MCAM) or cell surface glycoprotein MUC18, is a 113kDa cell adhesion molecule currently used as a marker for endothelial cell lineage. MCAM, a member of the immunoglobulin superfamily, is homologous to several cell adhesion molecules and is associated with tumor progression and the development of metastasis in human malignant melanoma. By radiation hybrid analysis, this gene is mapped to chromosome 11q23.3. MCAM has been demonstrated to appear on a small subset of T and B lymphocytes in the peripheral blood of healthy individuals. MCAM has been seen as a marker for mesenchymal stem cells isolated from multiple adult and fetal organs, and its expression may be linked to multipotency mesenchymal stem cells with greater differentiation potential express higher levels of MCAM on the cell surface.

Overview

Product Name	Anti-CD146/MCAM Antibody Picoband™ (monoclonal, 4C12)
Reactive Species	Human
Description	Boster Bio Anti-CD146/MCAM Antibody Picoband™ (monoclonal, 4C12) catalog # M01683-3. Tested in Flow Cytometry, IHC, IHC-F, WB applications. This antibody reacts with Human.
Application	Flow Cytometry, IHC, IHC-F, WB
Clonality	Monoclonal 4C12
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	Mouse
Uniprot ID	P43121

Technical Details

Immunogen	E.coli-derived human CD146 recombinant protein (Position: H59-A401). Human CD146 shares 73% amino acid (aa) sequence identity with both mouse and rat CD146.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Mouse IgG (EK1001) for Western blot, and HRP Conjugated anti-Mouse IgG Super Vision Assay Kit (SV0001-1) for IHC(P) and IHC(F).
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Mouse IgG2a
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</p> <p>Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human</p> <p>Immunohistochemistry (Frozen Section), 0.5-1ug/ml, Human</p> <p>Flow Cytometry, 1-3ug/1x10⁶ cells, Human</p>

Anti-CD146/MCAM Antibody Picoband™ (monoclonal, 4C12) (M01683-3) Images

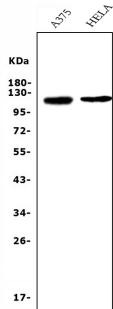


Figure 1. Western blot analysis of CD146/MCAM using anti-CD146/MCAM antibody (M01683-3). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human A375 whole cell lysates;
Lane 2: human Hela 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 mouse anti-CD146/MCAM antigen affinity purified monoclonal antibody (Catalog # M01683-3) 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-mouse 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 # EK1001) with Tanon 5200 system. A specific band was detected for CD146/MCAM at approximately 120KD. The expected band size for CD146/MCAM is at 72KD.

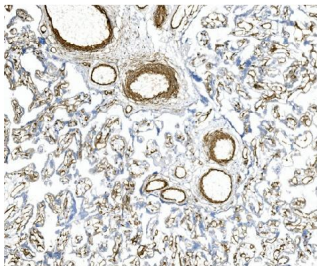


Figure 2. IHC analysis of CD146/MCAM using anti-CD146/MCAM antibody (M01683-3). CD146/MCAM was detected in paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-CD146/MCAM Antibody (M01683-3) overnight at 4°C. Biotinylated goat anti-mouse 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 # SA1021) with DAB as the chromogen.

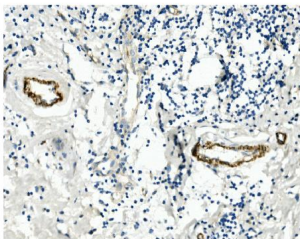
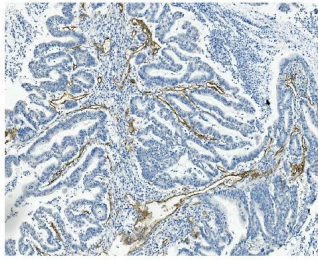


Figure 3. IHC analysis of CD146/MCAM using anti-CD146/MCAM antibody (M01683-3). CD146/MCAM was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-CD146/MCAM Antibody (M01683-3) overnight at 4°C. Biotinylated goat anti-mouse 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 # SA1021) with DAB as the chromogen.

Figure 4. IHC analysis of CD146/MCAM using anti-CD146/MCAM antibody (M01683-3).



CD146/MCAM was detected in paraffin-embedded section of human rectal cancer 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 mouse anti-CD146/MCAM Antibody (M01683-3) overnight at 4°C. Biotinylated goat anti-mouse 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 # SA1021) with DAB as the chromogen.

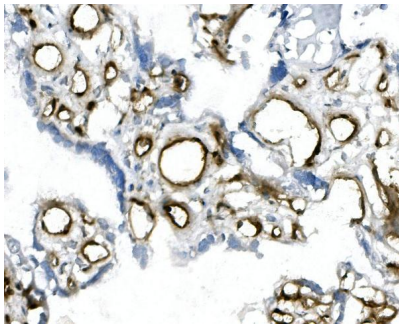


Figure 5. IHC analysis of CD146/MCAM using anti-CD146/MCAM antibody (M01683-3). CD146/MCAM was detected in frozen section of human placenta tissue. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-CD146/MCAM Antibody (M01683-3) overnight at 4°C. Biotinylated goat anti-mouse 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 # SA1021) with DAB as the chromogen.

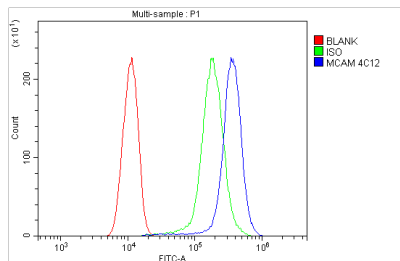


Figure 6. Flow Cytometry analysis of SiHa cells using anti-CD146/MCAM antibody (M01683-3). Overlay histogram showing SiHa cells stained with M01683-3 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-CD146/MCAM Antibody (M01683-3, 1ug/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10ug/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1ug/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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