

Anti-Methylmalonyl Coenzyme A mutase Antibody Picoband™

Catalog Number: A01065

About MUT

Methylmalonyl-CoA mutase (MUT) is a mitochondrial enzyme that catalyzes the isomerization of methylmalonyl-CoA to succinyl-CoA. This gene is mapped to 6p12.3. MUT requires a vitamin B12-derived prosthetic group, adenosylcobalamin (commonly referred to as AdoCbl), to function. And the product of this enzyme, succinyl-CoA, is a key molecule of the TCA cycle.

Overview

Product Name	Anti-Methylmalonyl Coenzyme A mutase Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-Methylmalonyl Coenzyme A mutase Antibody Picoband™ catalog # A01065. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 5mg BSA, 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	P22033

Technical Details

Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human MUT, different from the related mouse sequence by one amino acid.
Predicted Reactive Species	Human
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

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, Human, Mouse, Rat

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

Immunocytochemistry/Immunofluorescence, 2ug/ml, Human

Flow Cytometry, 1-3ug/1x10⁶ cells, Human

Anti-Methylmalonyl Coenzyme A mutase Antibody Picoband™ (A01065) Images

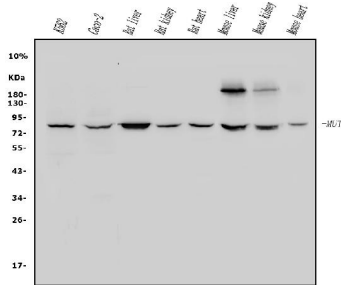


Figure 1. Western blot analysis of MUT using anti-MUT antibody (A01065).

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 K562 whole cell lysates,
lane 2: huam CACO-2 whole cell lysates,
lane 3: rat liver tissue lysates,
lane 4: rat kidney tissue lysates,
lane 5: rat heart tissue lysates,
lane 6: mouse liver tissue lysates,
lane 7: mouse kidney tissue lysates,
lane 8: mouse heart tissue 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-MUT antigen affinity purified polyclonal antibody (Catalog # A01065) 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 MUT at approximately 83KD. The expected band size for MUT is at 83KD.

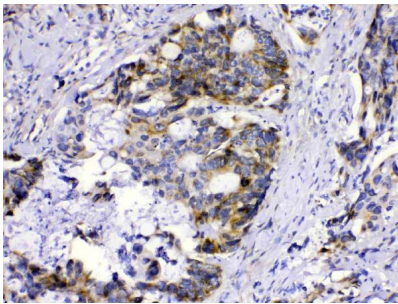


Figure 2. IHC analysis of MUT using anti-MUT antibody (A01065).

MUT was detected in paraffin-embedded section of human intestinal cancer 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-MUT Antibody (A01065) 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.

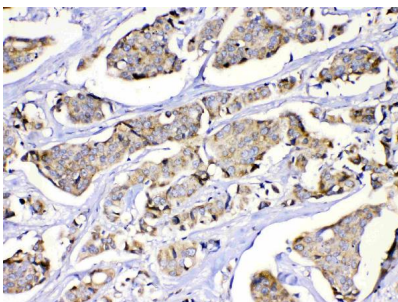


Figure 3. IHC analysis of MUT using anti-MUT antibody (A01065).

MUT was detected in paraffin-embedded section of human lung cancer 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-MUT Antibody (A01065) 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.

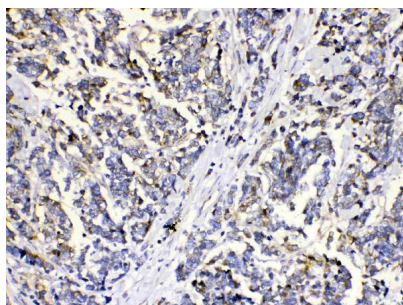


Figure 4. IHC analysis of MUT using anti-MUT antibody (A01065).

MUT was detected in paraffin-embedded section of human mammary cancer 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-MUT Antibody (A01065) 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.

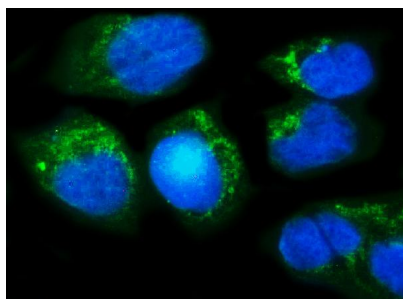


Figure 5. IF analysis of MUT using anti-MUT antibody (A01065).

MUT was detected in immunocytochemical section of U20S 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 2ug/mL rabbit anti-MUT Antibody (A01065) 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.

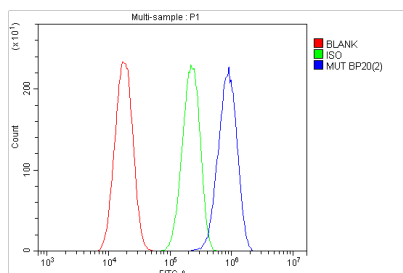


Figure 6. Flow Cytometry analysis of A431 cells using anti-MUT antibody (A01065).

Overlay histogram showing A431 cells stained with A01065 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-MUT Antibody (A01065, 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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