

Anti-MPI Picoband™ Antibody (monoclonal, 5G5)

Catalog Number: M00175-2

About MPI

Mannose-6 phosphate isomerase (MPI), alternately phosphomannose isomerase (PMI), is an enzyme which facilitates the interconversion of fructose 6-phosphate (F6P) and mannose-6-phosphate (M6P). It also plays a critical role in maintaining the supply of D-mannose derivatives, which are required for most glycosylation reactions. Mutations in the MPI gene were found in patients with carbohydrate-deficient glycoprotein syndrome, type Ib. Alternative splicing results in multiple transcript variants. This MPI gene is mapped to 15q24.1.

Overview

Product Name	Anti-MPI Picoband™ Antibody (monoclonal, 5G5)
Reactive Species	Human, Rat
Description	Boster Bio Anti-MPI Picoband™ Antibody (monoclonal, 5G5) catalog # M00175-2. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Rat.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Monoclonal 5G5
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na2HPO4.
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	P34949

Technical Details

Immunogen	E. coli-derived human MPI recombinant protein (Position: A2-K99). Human MPI shares 88.8% and 86.7% amino acid (aa) sequence identity with mouse and rat MPI, respectively.
Predicted Reactive Species	Hepatitis Virus
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 ICC.
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, 0.25-0.5ug/ml, Human, Rat</p> <p>Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human</p> <p>Immunocytochemistry/Immunofluorescence, 5ug/ml, Human</p> <p>Flow Cytometry, 1-3ug/1x10⁶ cells, Human</p>

Anti-MPI Picoband™ Antibody (monoclonal, 5G5) (M00175-2) Images

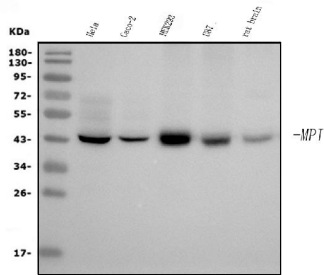


Figure 1. Western blot analysis of MPI using anti-MPI antibody (M00175-2).

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 50ug of sample under reducing conditions.

Lane 1: human HELA whole cell lysates,
Lane 2: human CACO-2 whole cell lysates,
Lane 3: human HEK293 whole cell lysates,
Lane 4: human U87 whole cell lysates,
Lane 5: rat brain 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 mouse anti-MPI antigen affinity purified monoclonal antibody (Catalog # M00175-2) 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-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 MPI at approximately 45KD. The expected band size for MPI is at 45KD.

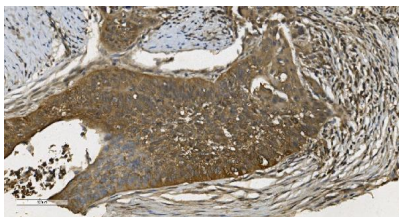


Figure 2. IHC analysis of MPI using anti-MPI antibody (M00175-2).

MPI was detected in paraffin-embedded section of human lung 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 2ug/ml mouse anti-MPI Antibody (M00175-2) 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.

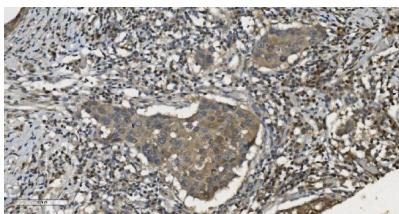
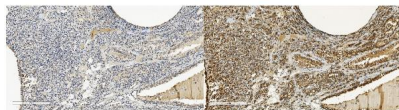


Figure 3. IHC analysis of MPI using anti-MPI antibody (M00175-2).

MPI was detected in paraffin-embedded section of human breast 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 2ug/ml mouse anti-MPI Antibody (M00175-2) 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 MPI using anti-MPI antibody



(M00175-2).

MPI was detected in paraffin-embedded section of human renal clear cell carcinoma 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 2ug/ml mouse anti-MPI Antibody (M00175-2) 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.

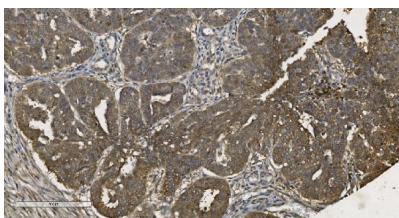


Figure 5. IHC analysis of MPI using anti-MPI antibody (M00175-2).

MPI was detected in paraffin-embedded section of human ovarian serous adenocarcinoma 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 2ug/ml mouse anti-MPI Antibody (M00175-2) 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.

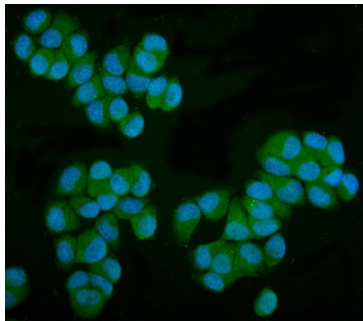


Figure 6. IF analysis of MPI using anti-MPI antibody (M00175-2).

MPI was detected in immunocytochemical section of T-47D 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 mouse anti-MPI Antibody (M00175-2) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) 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.

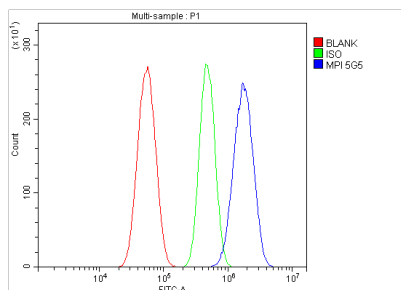


Figure 7. Flow Cytometry analysis of U87 cells using anti-MPI antibody (M00175-2).

Overlay histogram showing U87 cells stained with M00175-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-MPI Antibody (M00175-2, 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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