Anti-MTAP Antibody [JE63-74]

HA721955

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P, IF-Tissue, FC
Molecular Wt:	Predicted band size: 31 kDa
Clone number:	JE63-74

Description: S-methyl-5'-thioadenosine phosphorylase (MTAP) is an enzyme in humans responsible for polyamine metabolism. It is encoded by the methylthioadenosine phosphorylase (MTAP) gene on chromosome 9. Multiple alternatively spliced transcript variants have been described for this gene, but their full-length natures remain unknown. This gene encodes an enzyme that plays a major role in polyamine metabolism and is important for the salvage of both adenine and methionine. It is responsible for the first step in this pathway, where it catalyzes the reversible phosphorylation of MTA to adenine and 5-methylthioribose-1-phosphate. This takes place after MTA is generated from S-adenosylmethionine.

Immunogen: Recombinant protein within Human MTAP aa 184-283 / 283.

Positive control: HeLa cell lysate, NIH/3T3 cell lysate, C6 cell lysate, NIH/3T3, mouse liver tissue.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: Q13126 Human | Q9CQ65 Mouse
Entrez Gene: 298227 Rat

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:100
IHC-P	1:500
IF-Tissue	1:200
FC	1:1,000

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

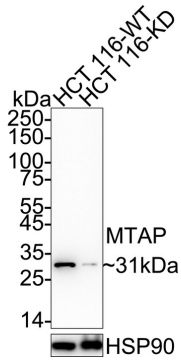

Fig1: Western blot analysis of MTAP on different lysates with Rabbit anti-MTAP antibody (HA721955) at 1/2,000 dilution.

Lane 1: HCT 116-si NT cell lysate
Lane 2: HCT 116-si MTAP cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 31 kDa
Observed band size: 31 kDa

Exposure time: 40 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA721955) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

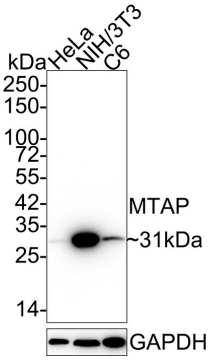

Fig2: Western blot analysis of MTAP on different lysates with Rabbit anti-MTAP antibody (HA721955) at 1/1,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: NIH/3T3 cell lysate
Lane 3: C6 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 31 kDa
Observed band size: 31 kDa

Exposure time: 24 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA721955) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

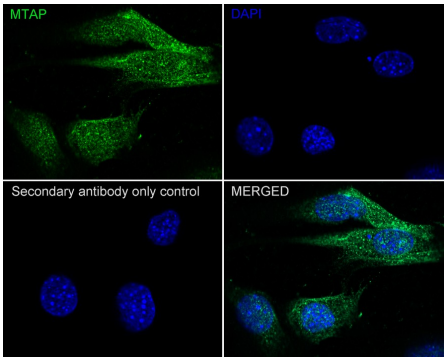

Fig3: Immunocytochemistry analysis of NIH/3T3 cells labeling MTAP with Rabbit anti-MTAP antibody (HA721955) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-MTAP antibody (HA721955) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

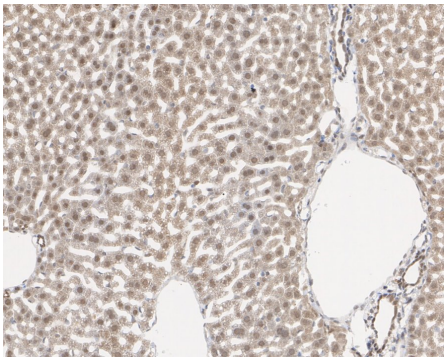

Fig4: Immunohistochemical analysis of paraffin-embedded mouse liver tissue with Rabbit anti-MTAP antibody (HA721955) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA721955) at 1/500 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

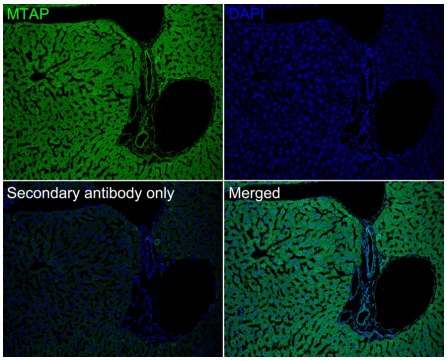

Fig5: Immunofluorescence analysis of paraffin-embedded mouse liver tissue labeling MTAP with Rabbit anti-MTAP antibody (HA721955) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA721955, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

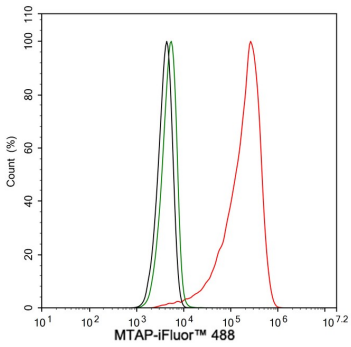

Fig6: Flow cytometric analysis of NIH/3T3 cells labeling MTAP.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA721955, 1μg/mL) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Kalev P et al. MAT2A Inhibition Blocks the Growth of MTAP-Deleted Cancer Cells by Reducing PRMT5-Dependent mRNA Splicing and Inducing DNA Damage. Cancer Cell. 2021 Feb
2. Hu Q et al. MTAP Deficiency-Induced Metabolic Reprogramming Creates a Vulnerability to Cotargeting De Novo Purine Synthesis and Glycolysis in Pancreatic Cancer. Cancer Res. 2021 Oct