

Anti-TPMT Antibody [JE64-09]

HA721049



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, FC
Molecular Wt:	Predicted band size: 28 kDa
Clone number:	JE64-09

Description: Thiopurine methyltransferase or thiopurine S-methyltransferase (TPMT) is an enzyme that in humans is encoded by the TPMT gene. A pseudogene for this locus is located on chromosome 18q. Thiopurine methyltransferase methylates thiopurine compounds. The methyl donor is S-adenosyl-L-methionine, which is converted to S-adenosyl-L-homocysteine. This enzyme metabolizes thiopurine drugs via S-adenosyl-L-methionine as the S-methyl donor and S-adenosyl-L-homocysteine as a byproduct. Thiopurine drugs such as 6-mercaptopurine are used as chemotherapeutic agents and immunosuppressive drugs. Genetic polymorphisms that affect this enzymatic activity are correlated with variations in sensitivity and toxicity to such drugs within individuals. About 1/300 individual is deficient for the enzyme. Genetic variants of TPMT have also been associated with cisplatin-induced ototoxicity in children. TPMT is now listed as a pharmacogenomic biomarker for adverse drug reactions to cisplatin by the FDA.

Immunogen: Synthetic peptide within human TPMT aa 101-150/245.

Positive control: K562 cell lysate, SK-Br-3 cell lysate, HepG2 cell lysate, A431 cell lysate, mouse brain tissue lysate, rat stomach tissue lysate, mouse kidney tissue lysate, rat lung tissue lysate, mouse hippocampus tissue, K562, mouse brain tissue.

Subcellular location: Cytoplasm.

Database links: SwissProt: P51580 Human | O55060 Mouse | Q9Z0T0 Rat

Recommended Dilutions:

WB	1:500
IHC-P	1:400-1:1,000
FC	1:500-1:1,000

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

Storage Instruction: Store at +4°C after thawing. Aliquot store at -20°C. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Hangzhou HuaAn Biotechnology Co.,Ltd.

Orders: 0086-571-88062880

Technical:0086-571-89986345

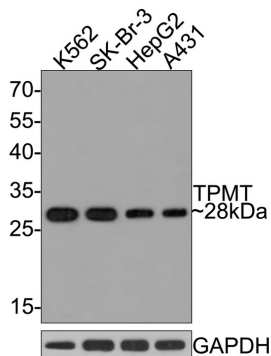
Service mail: support@huabio.cn

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Fig1: Western blot analysis of TPMT on different lysates with Rabbit anti-TPMT antibody (HA721049) at 1/500 dilution.

Lane 1: K562 cell lysate
 Lane 2: SK-Br-3 cell lysate
 Lane 3: HepG2 cell lysate
 Lane 4: A431 cell lysate



Lysates/proteins at 10 µg/Lane.

Predicted band size: 28 kDa
 Observed band size: 28 kDa

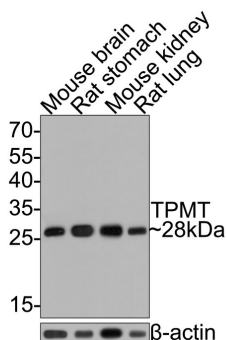
Exposure time: 2 minutes;

12% 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 (HA721049) at 1/500 dilution was used in 5% NFDm/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

Fig2: Western blot analysis of TPMT on different lysates with Rabbit anti-TPMT antibody (HA721049) at 1/500 dilution.

Lane 1: Mouse brain tissue lysate
 Lane 2: Rat stomach tissue lysate
 Lane 3: Mouse kidney tissue lysate
 Lane 4: Rat lung tissue lysate



Lysates/proteins at 20 µg/Lane.

Predicted band size: 28 kDa
 Observed band size: 28 kDa

Exposure time: 2 minutes;

12% 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 (HA721049) at 1/500 dilution was used in 5% NFDm/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

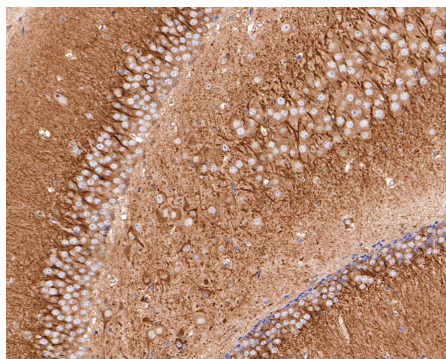


Fig3: Immunohistochemical analysis of paraffin-embedded mouse hippocampus tissue with Rabbit anti-TPMT antibody (HA721049) at 1/400 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA721049) at 1/400 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

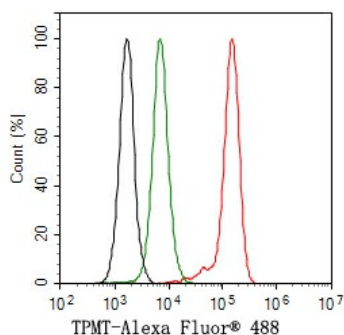


Fig4: Flow cytometric analysis of K562 cells labeling TPMT.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA721049, 1ug/ml) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C for an hour, the cells were stained with a Alexa Fluor® 488 conjugate-Goat anti-Rabbit IgG Secondary antibody at 1/1,000 dilution for 30 minutes at +4°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

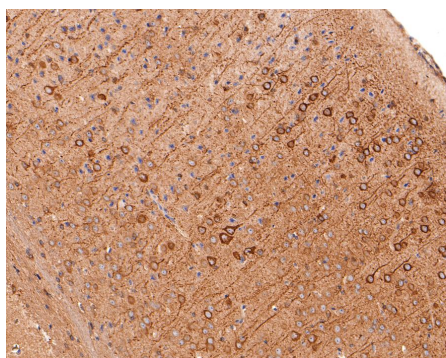


Fig5: Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Rabbit anti-TPMT antibody (HA721049) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA721049) at 1/1,000 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

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

Background References

1. Mhanna M. et. al. TPMT Genotype and Adverse Effects of Azathioprine among Jordanian Group. *Curr Drug Metab.* 2019
2. Harmand PO. et. al. Thiopurine Drugs in the Treatment of Ulcerative Colitis: Identification of a Novel Deleterious Mutation in TPMT. *Genes (Basel)*. 2020 Oct

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Applications: WB=Western blot IP=Immunoprecipitation IHC=Immunohistochemistry IF=Immunofluorescence FC=Flow cytometry