Anti-ENPP3 / B10 Antibody [PSH06-96]

HA722774

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 100 kDa
Clone number:	PSH06-96

Description: Ectonucleotide pyrophosphatase/phosphodiesterase family member 3 is an enzyme that in humans is encoded by the ENPP3 gene. The protein encoded by this gene belongs to a series of ectoenzymes that are involved in hydrolysis of extracellular nucleotides. These ectoenzymes possess ATPase and ATP pyrophosphatase activities and are type II transmembrane proteins. Expression of the related rat mRNA has been found in a subset of immature glial cells and in the alimentary tract. The corresponding rat protein has been detected in the pancreas, small intestine, colon, and liver. The human mRNA is expressed in glioma cells, prostate, and uterus. Expression of the human protein has been detected in uterus, basophils, and mast cells. This protein has also been used in conjunction with CD63 as a marker for activated basophils in the Basophil Activation Test for IgE mediated allergic reactions.

Immunogen: Recombinant protein within human ENPP3 aa 526-875.

Positive control: Mouse kidney tissue lysate, Rat uterus tissue lysate, Rat kidney tissue lysate, Rat liver tissue lysate, 293T transfected with ENPP3 cell lysate, HeLa cells transfected with ENPP3, human endometrium tissue, mouse endometrium tissue.

Subcellular location: Cell membrane, Apical cell membrane, Secreted.

Database links: SwissProt: O14638 Human | Q6DYE8 Mouse | P97675 Rat

Recommended Dilutions:

WB	1:1,000-1:2,000
IF-Cell	1:100
IHC-P	1:200-1:1,000

Storage Buffer: PBS (pH7.4), 0.1% 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

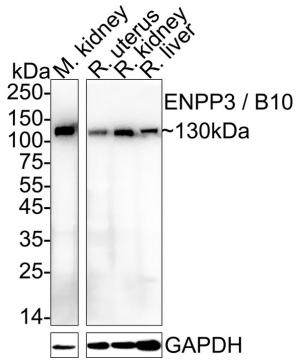

Fig1: Western blot analysis of ENPP3 / B10 on different lysates with Rabbit anti-ENPP3 / B10 antibody (HA722774) at 1/1,000 dilution.

Lane 1: Mouse kidney tissue lysate
Lane 2: Rat uterus tissue lysate
Lane 3: Rat kidney tissue lysate
Lane 4: Rat liver tissue lysate

Lysates/proteins at 40 µg/Lane.

Predicted band size: 100 kDa
Observed band size: 130 kDa

Exposure time: 25 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 (HA722774) 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.

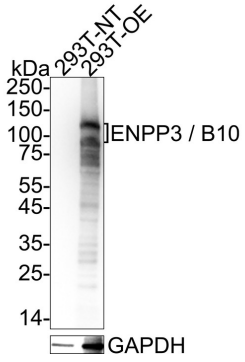

Fig2: Western blot analysis of ENPP3 / B10 on different lysates with Rabbit anti-ENPP3 / B10 antibody (HA722774) at 1/2,000 dilution.

Lane 1: 293T-NT cell lysate
Lane 2: 293T transfected with ENPP3 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 100 kDa
Observed band size: 100/130 kDa

Exposure time: 4 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 (HA722774) 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.

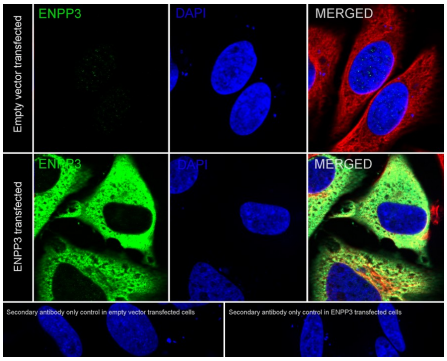

Fig3: Immunocytochemistry analysis of HeLa cells transfected with or without ENPP3 labeling ENPP3 / B10 with Rabbit anti-ENPP3 / B10 antibody (HA722774) at 1/100 dilution.

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-ENPP3 / B10 antibody (HA722774) 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.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

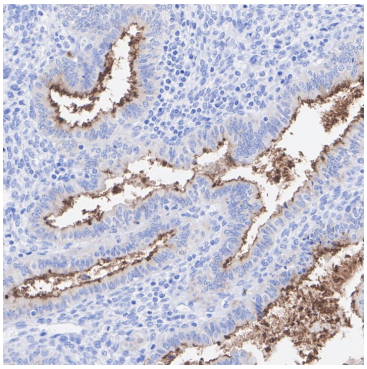

Fig4: Immunohistochemical analysis of paraffin-embedded human endometrium tissue with Rabbit anti-ENPP3 / B10 antibody (HA722774) at 1/1,000 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (HA722774) 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.

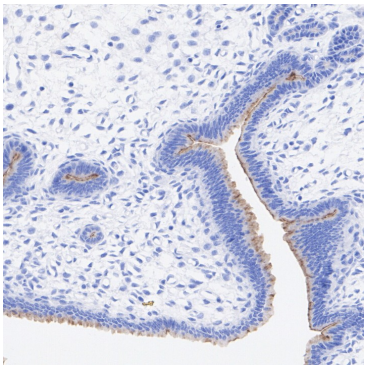

Fig5: Immunohistochemical analysis of paraffin-embedded mouse endometrium tissue with Rabbit anti-ENPP3 / B10 antibody (HA722774) at 1/1,000 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (HA722774) 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.

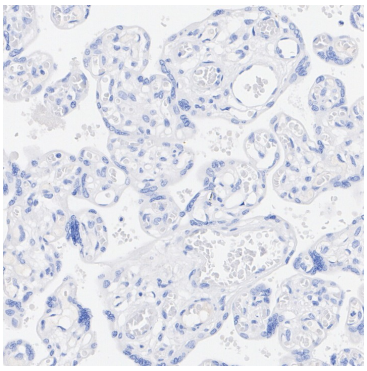

Fig6: Immunohistochemical analysis of paraffin-embedded human placenta tissue (negative) with Rabbit anti-ENPP3 / B10 antibody (HA722774) at 1/1,000 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (HA722774) 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.

Background References

1. Qin Y et al. Hypomethylation of the ENPP3 promoter region contributes to the occurrence and development of ovarian endometriosis via the AKT/mTOR/4EBP1 signaling pathway. Biomol Biomed. 2023 Dec
2. Zhou M et al. GRIA2/ENPP3 Regulates the Proliferation and Migration of Vascular Smooth Muscle Cells in the Restenosis Process Post-PTA in Lower Extremity Arteries. Front Physiol. 2021 Aug