# **Anti-PTEN Antibody [SJ19-03]**

## ET1606-43



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P
Molecular Wt:	Predicted band size: 47 kDa
Clone number:	SJ19-03
Description:	As human tumors progress to advanced stages, one genetic alteration that occurs at high frequency is a loss of heterozygosity (LOH) at chromosome 10q23. Mapping of homozygous deletions on this chromosome led to the isolation of the PTEN gene, also designated MMAC1 (for mutated in multiple advanced cancers) and TEP1. This candidate tumor suppresson gene exhibits a high frequency of mutations in human glioblastomas and is also mutated in other cancers, including sporadic brain, breast, kidney and prostate cancers. PTEN has been associated with Cowden disease, an autosomal dominant cancer predisposition syndrome. The PTEN gene product is a putative protein tyrosine phosphatase that is localized to the cytoplasm and shares extensive homology with the cytoskeletal proteins tensin and auxilin. Gene transfer studies have indicated that the phosphatase domain or PTEN is essential for growth suppression of glioma cells.
lmmunogen:	Recombinant full length protein of human PTEN.
Positive control:	MCF7 cell lysate, A431 cell lysate, HeLa cell lysate, NIH/3T3 cell lysate, C6 cell lysate, PC- 12 cell lysate, NIH/3T3, mouse brain tissue, mouse pancreas tissue, rat brain tissue, rat pancreas tissue.
Subcellular location:	Cytoplasm, Nucleus, Secreted.
Database links:	SwissProt: P60484 Human   O08586 Mouse Unigene: 22158 Rat
Recommended Dilutions: WB IF-Cell IF-Tissue IHC-P	1:1,000-1:5,000 1:100-1:500 1:50 1:200
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!C$ after thawing. Aliquot store at -20 $^\circ\!C$ or -80 $^\circ\!C$ . Avoid repeated freeze / thaw cycles.

# Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

#### Images

ET1606-43

1/2,000

250 150

14

Competitor C

1/1,000

250 150

100-72-55-42-35-

25-

14

10

**Fig1:** Western blot analysis of PTEN on different lysates with Rabbit anti-PTEN antibody (ET1606-43) at 1/2,000 dilution and competitor's antibody at 1/1,000 dilution.

Lane 1: MCF7 cell lysate Lane 2: MDA-MB-468 cell lysate (negative) Lane 3: A431 cell lysate Lane 4: HeLa cell lysate Lane 5: NIH/3T3 cell lysate Lane 6: C6 cell lysate Lane 7: PC-12 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 47 kDa Observed band size: 55 kDa

Exposure time: 1 minute 10 seconds; ECL: K1802; 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 (ET1606-43) at 1/2,000 dilution and competitor's antibody at 1/1,000 dilution were used in 5% NFDM/TBST at 4°C 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 PTEN on different lysates with Rabbit anti-PTEN antibody (ET1606-43) at 1/1,000 dilution.

Lane 1: A549-si NT cell lysate Lane 2: A549-si PTEN cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 47 kDa Observed band size: 55 kDa

Exposure time: 45 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 (ET1606-43) at 1/1,000 dilution was used in 5% NFDM/TBST at  $4^{\circ}$ C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

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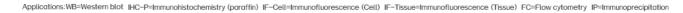


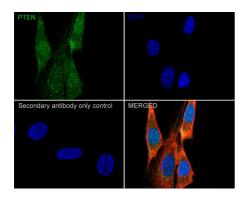
35-25-14-\_\_\_\_\_ GAPDH

45

PTEN

~55kDa

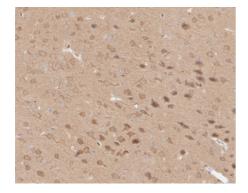




**Fig3:** Immunocytochemistry analysis of NIH/3T3 cells labeling PTEN with Rabbit anti-PTEN antibody (ET1606-43) at 1/100 dilution.

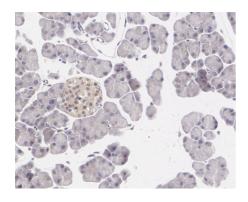
Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-PTEN antibody (ET1606-43) at 1/100 dilution in 1% BSA in PBST overnight at 4  $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor<sup>TM</sup> 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  $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor 150 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.



**Fig4:** Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Rabbit anti-PTEN antibody (ET1606-43) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1606-43) at 1/200 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 pancreas tissue with Rabbit anti-PTEN antibody (ET1606-43) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1606-43) at 1/200 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.

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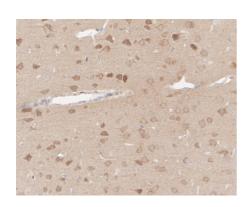
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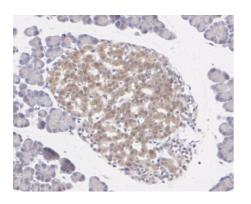


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**Fig6:** Immunohistochemical analysis of paraffin-embedded rat brain tissue with Rabbit anti-PTEN antibody (ET1606-43) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1606-43) at 1/200 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.



**Fig7:** Immunohistochemical analysis of paraffin-embedded rat pancreas tissue with Rabbit anti-PTEN antibody (ET1606-43) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1606-43) at 1/200 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. Chen J et al. Low expression of phosphatase and tensin homolog in clear-cell renal cell carcinoma contributes to chemoresistance through activating the Akt/HDM2 signaling pathway. Mol Med Rep 12:2622-8 (2015).
- Zeng C et al. SPOP suppresses tumorigenesis by regulating hedgehog/Gli2 signaling pathway in gastric cancer. J Exp Clin Cancer Res 33:75 (2014).

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