

Anti-PEN2 Antibody [JE41-28]

ET7109-26



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P, FC, IHC-Fr, IF-Tissue
Molecular Wt:	Predicted band size: 12 kDa
Clone number:	JE41-28

Description: Four proteins comprise the gamma-secretase complex: presenilin, nicastrin, aph-1, and PEN-2. Together, these proteins mediate cell surface signaling pathways for a variety of type I membrane proteins, notably amyloid-beta precursor protein, a protein implicated in the development of Alzheimer's disease, via intramembrane proteolysis. The proteins assemble into a proteolytically active complex in the golgi/trans-golgi network (TGN) compartments. Assembly leads to autocleavage of presenilin into two subunits to create the active site of gamma-secretase, an important step in understanding the mechanisms involved in the etiology and possible treatment of Alzheimer's disease.

Immunogen: Recombinant protein within Human PEN2 aa 1-101 / 101.

Positive control: Mouse spleen tissue lysates, A549, SH-SY5Y, human lung carcinoma tissue, human kidney tissue, rat cerebellum tissue, mouse testis tissue, human tonsil tissue.

Subcellular location: Cell membrane. Endoplasmic reticulum. Golgi apparatus.

Database links: SwissProt: Q9NZ42 Human | Q9CQR7 Mouse | O88777 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IHC-P	1:50-1:200
IF-Cell	1:50-1:200
FC	1:50-1:100
IHC-Fr	1:100
IF-Tissue	1:200

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.

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Images

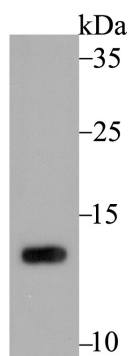


Fig1: Western blot analysis of PEN2 on mouse spleen tissue lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET7109-26, 1/2,000) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

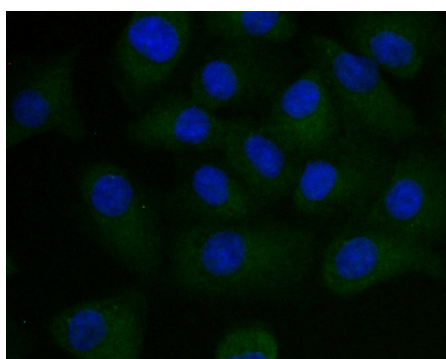


Fig2: ICC staining of PEN2 in A549 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET7109-26, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

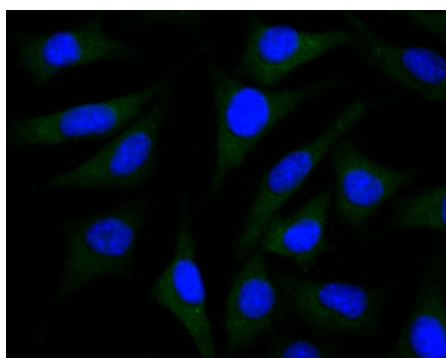


Fig3: ICC staining of PEN2 in SH-SY5Y cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET7109-26, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

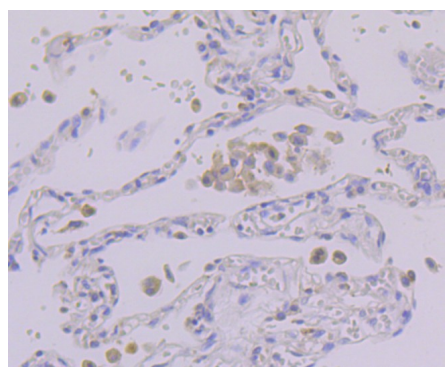


Fig4: Immunohistochemical analysis of paraffin-embedded human lung carcinoma tissue using anti-PEN2 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET7109-26, 1/50) for 30 minutes 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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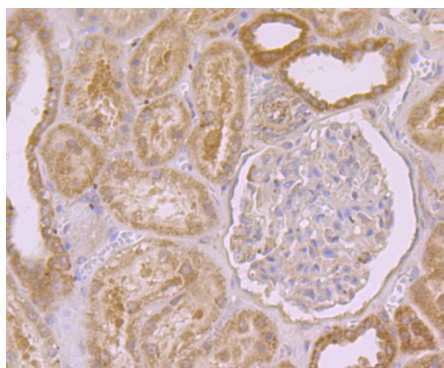


Fig5: Immunohistochemical analysis of paraffin-embedded human kidney tissue using anti-PEN2 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET7109-26, 1/50) for 30 minutes 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.

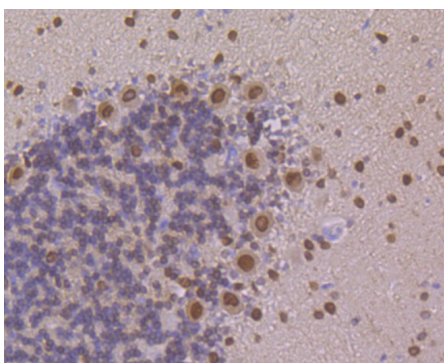


Fig6: Immunohistochemical analysis of paraffin-embedded rat cerebellum tissue using anti-PEN2 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET7109-26, 1/50) for 30 minutes 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.

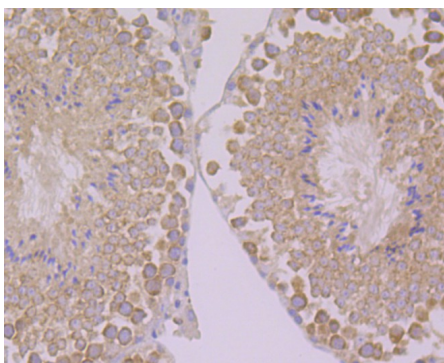


Fig7: Immunohistochemical analysis of paraffin-embedded mouse testis tissue using anti-PEN2 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET7109-26, 1/50) for 30 minutes 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.

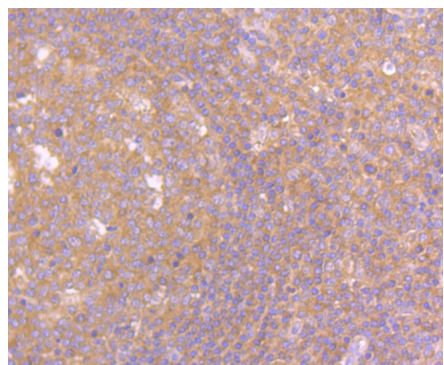


Fig8: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-PEN2 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET7109-26, 1/50) for 30 minutes 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.

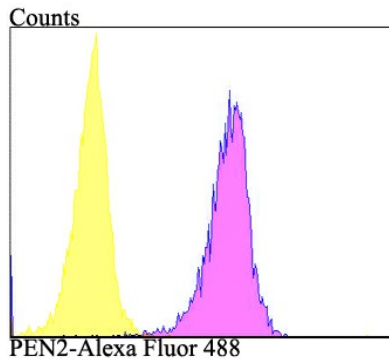


Fig9: Flow cytometric analysis of PEN2 was done on A549 cells. The cells were fixed, permeabilized and stained with the primary antibody (ET7109-26, 1/50) (purple). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes. Unlabelled sample was used as a control (cells without incubation with primary antibody; yellow).

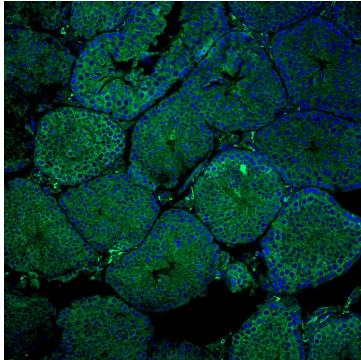


Fig10: Immunofluorescence analysis of frozen mouse testis tissue with Rabbit anti-PEN2 antibody (ET7109-26) at 1/100 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. 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 (ET7109-26, green) at 1/100 dilution overnight at 4 °C, 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).

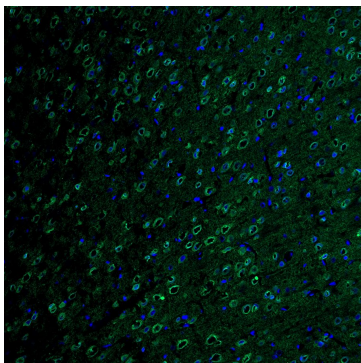


Fig11: Immunofluorescence analysis of frozen rat brain tissue with Rabbit anti-PEN2 antibody (ET7109-26) at 1/100 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. 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 (ET7109-26, green) at 1/100 dilution overnight at 4 °C, 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).

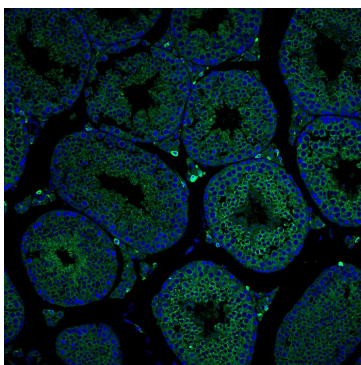


Fig12: Immunofluorescence analysis of paraffin-embedded mouse testis tissue labeling PEN2 with Rabbit anti-PEN2 antibody (ET7109-26) at 1/200 dilution.

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 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (ET7109-26, green) at 1/200 dilution overnight at 4 °C, 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).

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

Background References

1. Luo W-J et al. PEN-2 and APH-1 coordinately regulate proteolytic processing of presenilin 1. *J Biol Chem* 278:7850-7854 (2003).
2. Marlow L et al. APH1, PEN2, and nicastrin increase Abeta levels and gamma-secretase activity. *Biochem Biophys Res Commun* 305:502-509 (2003).

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