Anti-Neuropilin 1 Antibody [ST05-30]

ET1609-69



Product Type: Recombinant Rabbit monoclonal IgG, primary antibodies

Species reactivity: Human, Mouse, Rat

Applications: WB, IF-Cell, IF-Tissue, IHC-P, IP

Molecular Wt: Predicted band size: 103 kDa

Clone number: ST05-30

Description: Neuropilin is a type I transmembrane receptor that has been implicated in aspects of axon

growth and guidance and has been shown to act as a high affinity receptor for class III semaphorins and vascular endothelial growth factor (VEGF). A closely related protein, neuropilin-2, shares a common domain structure and significant homology with neuropilin and also acts as a receptor for the class III semaphorins and VEGF. Both neuropilins are involved in regulating many physiological pathways including axonal guidance and angiogenesis, however they exhibit differential expression in the adult vasculature. Neuropilin-2 is polysialylated and expressed on the surface of dendritic cells. It is also expressed by venous and lymphatic endothelium. Neuropilin is expressed predominantly by

arterial endothelium.

Immunogen: Synthetic peptide within Human Neuropilin-1 aa 881-923 / 923.

Positive control: U-87 MG cell lysate, A549 cell lysate, MDA-MB-231 cell lysate, mouse brain tissue lysate,

rat brain tissue lysate, human liver tissue, mouse heart tissue lysate, rat heart tissue lysate,

SHG-44, MCF-7, HUVEC, human kidney tissue, mouse kidney tissue.

Subcellular location: Cell membrane, Secreted.

Database links: SwissProt: O14786 Human | P97333 Mouse | Q9QWJ9 Rat

Recommended Dilutions:

WB 1:2,000-1:5,000
IF-Cell 1:50-1:200
IF-Tissue 1:50-1:200
IHC-P 1:50-1:200

IP Use at an assay dependent concentration.

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

Storage Instruction: Shipped at 4° C. Store at $+4^{\circ}$ C short term (1-2 weeks). It is recommended to aliquot into

single-use upon delivery. Store at -20°C long term.

Purity: Protein A affinity purified.

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Images

Fig1: Western blot analysis of Neuropilin 1 on different lysates with Rabbit anti-Neuropilin 1 antibody (ET1609-69) at 1/2,000 dilution and competitor's antibody at 1/2,000 dilution.

Lane 1: U-87 MG cell lysate Lane 2: A549 cell lysate

Lane 3: SK-Br-3 cell lysate (low expression)

Lane 4: MDA-MB-231 cell lysate Lane 5: Mouse brain tissue lysate Lane 6: Rat brain tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 103 kDa Observed band size: 130 kDa

Exposure time: 35 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 (ET1609-69) at 1/2,000 dilution and competitor's antibody at 1/2,000 dilution were used in 5% BSA at $4\,^{\circ}\mathrm{C}$ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

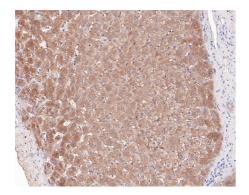


Fig2: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-Neuropilin 1 antibody (ET1609-69) at 1/200 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1609-69) 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.



Fig3: Western blot analysis of Neuropilin 1 on different lysates with Rabbit anti-Neuropilin 1 antibody (ET1609-69) at 1/5,000 dilution.

Lane 1: A549 cell lysate

Lane 2: MDA-MB-231 cell lysate

Lane 3: SK-Br-3 cell lysate (low expression)

Lane 4: Mouse brain tissue lysate Lane 5: Mouse heart tissue lysate Lane 6: Rat brain tissue lysate

Lane 7: Rat heart tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 103 kDa Observed band size: 130 kDa

Exposure time: Lane 1-3: 1 minute 21 seconds; Lane 4-7: 43

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 (ET1609-69) at 1/5,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:100,000 dilution was used for 1 hour at room temperature.

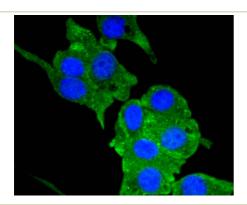


Fig4: ICC staining of Neuropilin 1 in SHG-44 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-69, 1/50) for 1 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).

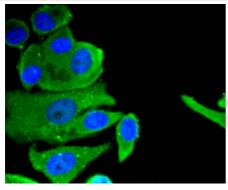


Fig5: ICC staining of Neuropilin 1 in MCF-7 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-69, 1/50) for 1 hour 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).

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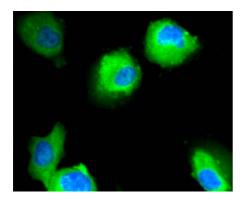


Fig6: ICC staining of Neuropilin 1 in HUVEC cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-69, 1/50) for 1 hour 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).

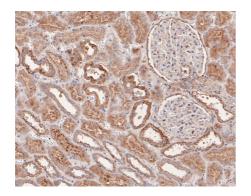


Fig7: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-Neuropilin 1 antibody (ET1609-69) at 1/50 dilution.

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 (ET1609-69) at 1/50 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.

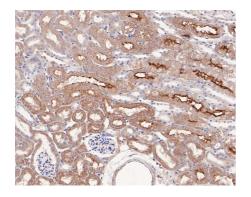


Fig8: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-Neuropilin 1 antibody (ET1609-69) at 1/200 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1609-69) 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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GAPDH

Fig9: Western blot analysis of Neuropilin 1 on different lysates with Rabbit anti-Neuropilin 1 antibody (ET1609-69) at 1/5,000 dilution.

Lane 1: MDA-MB-231-si NT cell lysate

Lane 2: MDA-MB-231-si Neuropilin 1 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 103 kDa Observed band size: 130 kDa

Exposure time: 1 minute 2 seconds;

4-20% SDS-PAGE gel.

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

Background References

- 1. Liu F et al. Prostate cancer cells induce osteoblastic differentiation via semaphorin 3A. Prostate 75:370-80 (2015).
- 2. Linthicum FH et al. The periductal channels of the endolymphatic duct, hydrodynamic implications. Otolaryngol Head Neck Surg 150:441-7 (2014).