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Anti-Neuropilin 1 Antibody Picoband™ (monoclonal, 4G3F7)

Catalog # ABO16266

Product Information

Application	WB, IF, ICC, FC
Primary Accession	O14786
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Rat, Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-Neuropilin 1 Antibody Picoband™ (monoclonal, 4G3F7) . Tested in Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Human, Rat.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 □g/ml.

Additional Information

Gene ID	8829
Other Names	Neuropilin-1, Vascular endothelial cell growth factor 165 receptor, CD304, NRP1 (HGNC:8004), NRP, VEGF165R
Calculated MW	103134
Application Details	Western blot, 0.25-0.5 □g/ml, Human, Rat Immunocytochemistry/Immunofluorescence, 5 □g/ml, Human Flow Cytometry, 1-3 □g/1x10^6 cells, Human
Source	Eukaryota
Contents	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.
Clone Names	Clone: 4G3F7
Immunogen	E.coli-derived human Neuropilin 1 recombinant protein (Position: K504-T827). Human Neuropilin 1 shares 95% and 94% amino acid (aa) sequences identity with mouse and rat Neuropilin 1, respectively.
Purification	Immunogen affinity purified.
Storage	At -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freezing and thawing.

Protein Information

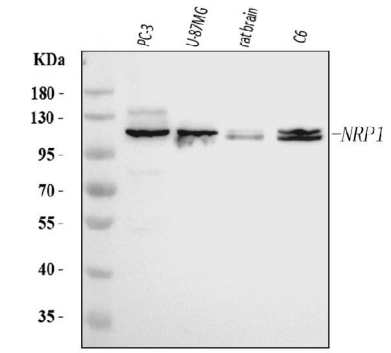
Name	NRP1 (HGNC:8004)
Synonyms	NRP, VEGF165R
Function	Cell-surface receptor involved in the development of the cardiovascular system, in angiogenesis, in the formation of certain neuronal circuits and in organogenesis outside the nervous system. Mediates the chemorepulsant activity of semaphorins (PubMed:10688880, PubMed:9288753, PubMed:9529250). Recognizes a C-end rule (CendR) motif R/KXXR/K on its ligands which causes cellular internalization and vascular leakage (PubMed:19805273). It binds to semaphorin 3A, the PLGF-2 isoform of PGF, the VEGF165 isoform of VEGFA and VEGFB (PubMed:10688880, PubMed:19805273, PubMed:9288753, PubMed:9529250). Coexpression with KDR results in increased VEGF165 binding to KDR as well as increased chemotaxis. Regulates VEGF-induced angiogenesis. Binding to VEGFA initiates a signaling pathway needed for motor neuron axon guidance and cell body migration, including for the caudal migration of facial motor neurons from rhombomere 4 to rhombomere 6 during embryonic development (By similarity). Regulates mitochondrial iron transport via interaction with ABCB8/MITOSUR (PubMed:30623799).
Cellular Location	[Isoform 2]: Secreted Secreted. Note=(Microbial infection) Proteolytically cleaved at the cell membrane upon H.pylori infection.
Tissue Location	[Isoform 1]: The expression of isoforms 1 and 2 does not seem to overlap. Expressed in olfactory epithelium (at protein level) (PubMed:33082293). Expressed in fibroblasts (at protein level) (PubMed:36213313). Expressed by the blood vessels of different tissues In the developing embryo it is found predominantly in the nervous system. In adult tissues, it is highly expressed in heart and placenta; moderately in lung, liver, skeletal muscle, kidney and pancreas; and low in adult brain (PubMed:10688880, PubMed:9529250). Expressed in the central nervous system, including olfactory related regions such as the olfactory tubercles and paraolfactory gyri (PubMed:33082293)

Background

This gene encodes one of two neuropilins, which contain specific protein domains which allow them to participate in several different types of signaling pathways that control cell migration. Neuropilins contain a large N-terminal extracellular domain, made up of complement-binding, coagulation factor V/VIII, and meprin domains. These proteins also contain a short membrane-spanning domain and a small cytoplasmic domain. Neuropilins bind many ligands and various types of co-receptors; they affect cell survival, migration, and attraction. Some of the ligands and co-receptors bound by neuropilins are vascular endothelial growth factor (VEGF) and semaphorin family members. Several alternatively spliced transcript variants that encode different protein isoforms have been described for this gene.

Images

Figure 1. Western blot analysis of Neuropilin 1 using anti-Neuropilin 1 antibody (M01324-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours.
The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human PC-3 whole cell lysates,
Lane 2: human U-87MG whole cell lysates,



Lane 3: rat brain tissue lysates,
Lane 4: rat C6 whole cell lysates.
After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-Neuropilin 1 antigen affinity purified monoclonal antibody (Catalog # M01324-1) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for Neuropilin 1 at approximately 120 kDa. The expected band size for Neuropilin 1 is at 120 kDa.

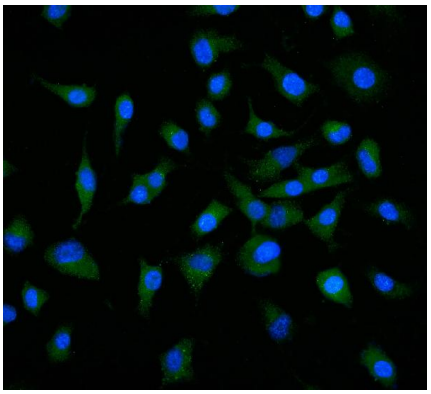


Figure 2. IF analysis of Neuropilin 1 using anti-Neuropilin 1 antibody (M01324-1).
Neuropilin 1 was detected in an immunocytochemical section of A549 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-Neuropilin 1 Antibody (M01324-1) overnight at 4°C. DyLight®488 Conjugated Goat Anti-mouse IgG (BA1126) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

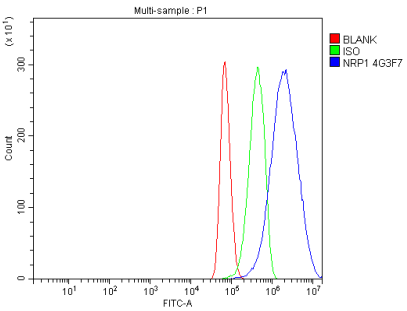


Figure 3. Flow Cytometry analysis of U87 cells using anti-Neuropilin 1 antibody (M01324-1).
Overlay histogram showing U87 cells stained with M01324-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Neuropilin 1 Antibody (M01324-1, 1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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