NF-L Recombinant Antibody [PS02-10] - Rat IgG1 (Chimeric)

HA601409

Product Type:	Recombinant Chimeric Antibody, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	IHC-Fr, IHC-P, WB
Molecular Wt:	Predicted band size: 62 kDa
Clone number:	PS02-10

Description: Neurofilament light polypeptide, also known as neurofilament light chain, abbreviated to NF-L or Nfl and with the HGNC name NEFL is a member of the intermediate filament protein family. This protein family consists of over 50 human proteins divided into 5 major classes, the Class I and II keratins, Class III vimentin, GFAP, desmin and the others, the Class IV neurofilaments and the Class V nuclear lamins. There are four major neurofilament subunits, NF-L, NF-M, NF-H and α-internexin. These form heteropolymers which assemble to produce 10nm neurofilaments which are only expressed in neurons where they are major structural proteins, particularly concentrated in large projection axons. Axons are particularly sensitive to mechanical and metabolic compromise and as a result axonal degeneration is a significant problem in many neurological disorders. The detection of neurofilament subunits in CSF and blood has therefore become widely used as a biomarker of ongoing axonal compromise. The NF-L protein is encoded by the NEFL gene. Neurofilament light chain is a biomarker that can be measured with immunoassays in cerebrospinal fluid and plasma and reflects axonal damage in a wide variety of neurological disorders. It is a useful marker for disease monitoring in amyotrophic lateral sclerosis, multiple sclerosis, Alzheimer's disease, and more recently Huntington's disease. It is also promising marker for follow-up of patients with brain tumors. Higher levels of blood or CSF NF-L have been associated with increased mortality, as would be expected as release of this protein reflects ongoing axonal loss. Recent work performed as a collaboration between EnCor Biotechnology Inc. and the University of Florida showed that the NF-L antibodies employed in the most widely used NF-L assays are specific for cleaved forms of NF-L generated by proteolysis induced by cell death.

Immunogen:	Recombinant protein.
Positive control:	Human cerebellum tissue, mouse cerebellum tissue, rat cerebellum tissue, Mouse brain tissue lysate, Rat brain tissue lysate.
Subcellular location:	Cell projection, axon, Cytoplasm, cytoskeleton.
Database links:	SwissProt: P07196 Human | P08551 Mouse | P19527 Rat
Recommended Dilutions:	
IHC-Fr	1:500
IHC-P	1:2,000-1:4,000
WB	1:2,000
Storage Buffer:	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

Images

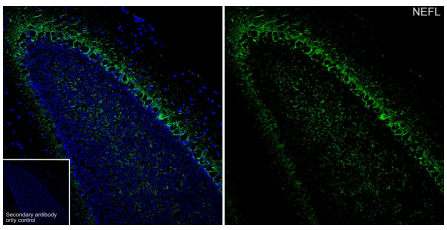

Fig1: Immunofluorescence analysis of frozen mouse cerebellum tissue with Rat anti-NF-L antibody (HA601409) at 1/500 dilution.

The section was not undergone antigen retrieval.

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 (HA601409, green) at 1/500 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rat IgG H&L (iFluor™ 488, HA1133) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

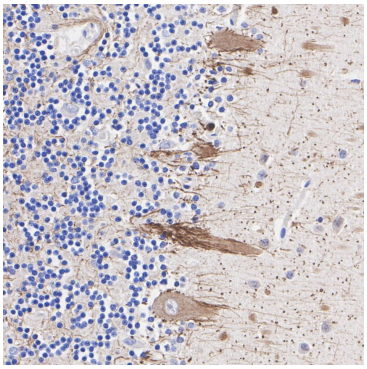

Fig2: Immunohistochemical analysis of paraffin-embedded human cerebellum tissue with Rat anti-NF-L antibody (HA601409) at 1/2,000 dilution.

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 (HA601409) at 1/2,000 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.

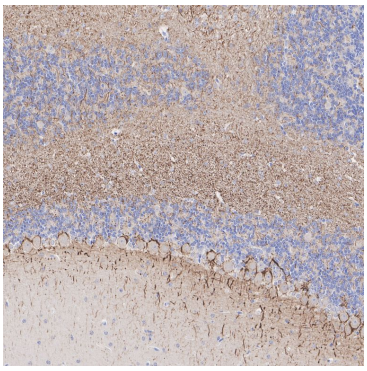

Fig3: Immunohistochemical analysis of paraffin-embedded mouse cerebellum tissue with Rat anti-NF-L antibody (HA601409) at 1/4,000 dilution.

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 (HA601409) at 1/4,000 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.

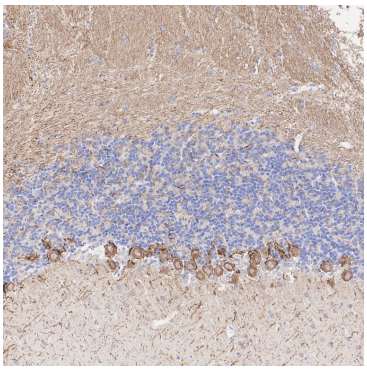

Fig4: Immunohistochemical analysis of paraffin-embedded rat cerebellum tissue with Rat anti-NF-L antibody (HA601409) at 1/4,000 dilution.

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 (HA601409) at 1/4,000 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.

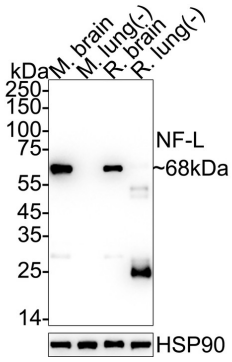

Fig5: Western blot analysis of NF-L on different lysates with Rat anti-NF-L antibody (HA601409) at 1/2,000 dilution.

Lane 1: Mouse brain tissue lysate
Lane 2: Mouse lung tissue lysate (negative)
Lane 3: Rat brain tissue lysate
Lane 4: Rat lung tissue lysate (negative)

Lysates/proteins at 20 µg/Lane.

Predicted band size: 62 kDa
Observed band size: 68 kDa

Exposure time: 10 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 (HA601409) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rat IgG H&L - HRP Secondary Antibody (HA1023) at 1/5,000 dilution was used for 1 hour at room temperature.

Background References

1. Gong L et al. Neurofilament Light Chain (NF-L) Stimulates Lipid Peroxidation to Neuronal Membrane through Microglia-Derived Ferritin Heavy Chain (FTH) Secretion. Oxid Med Cell Longev. 2022 Mar
2. Heiskanen M et al. Plasma Neurofilament Light Chain (NF-L) Is a Prognostic Biomarker for Cortical Damage Evolution but Not for Cognitive Impairment or Epileptogenesis Following Experimental TBI. Int J Mol Sci. 2022 Dec