Anti-Human/Mouse/Rat NF-L Antibody [PSH11-41] - BSA and Azide free (Capture)

HA723330

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	ELISA(Cap)
Clone number:	PSH11-41

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 within Human NF-L protein aa 62-407.
Positive control:	Recombinant Human NF-L protein.
Subcellular location:	Cell projection, axon, Cytoplasm, cytoskeleton.
Database links:	SwissProt: P07196 Human | P08551 Mouse | P19527 Rat
Recommended Dilutions:	
ELISA(Cap)	Use at an assay dependent concentration. Can be paired for Sandwich ELISA with Rabbit monoclonal [PSH11-42] to Human NF-L antibody (Detector) (HA723331) and Recombinant Human NF-L protein as the standard. The reference range value is 78-10,000 pg/ml.
Storage Buffer:	PBS (pH7.4).
Storage Instruction:	Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

Images

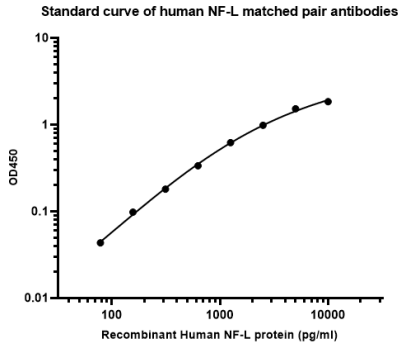

Fig1: Standard curve of human NF-L matched pair antibodies

Sandwich ELISA analysis of human NF-L matched pair antibodiesElisa assay was performed by coating wells of a 96-well plate with 100 µl per well of capture antibody (HA723330) diluted in carbonate/bicarbonate buffer, at a concentration of 2ug/ml overnight at 4℃. Wells of the plate were washed, blocked with 150 µl 0.05% tween-20 1% BSA blocking buffer, and incubated with serial diluted Recombinant Human NF-L protein starting from 10,000 pg/ml to 0 pg/ml and detect antibody (HA723331, Biotin, 0.2 µg/ml) for 1 hour at 30℃ with shaking. Then the plate was washed and incubated with 100 µl per well of SA-HRP for 0.5 hour at 30℃ with shaking. Detection was performed using an Ultra TMB Substrate for 10 minutes at room temperature in the dark. The reaction was stopped with sulfuric acid and absorbances were read on a spectrophotometer at 450 nm.

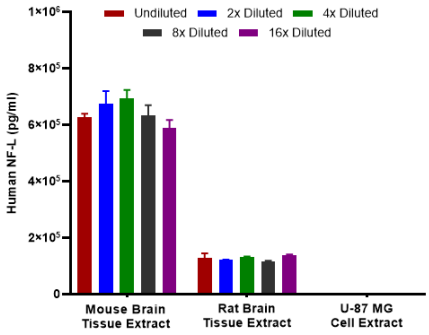

Fig2: Interpolated concentrations of native NF-L in mouse brain, rat brain and U-87 MG extract samples based on a 1000 µg/ml extract load.

Interpolated concentration of native NF-L was measured in duplicate at different sample concentrations and interpolated from the NF-L standard curves. The interpolated dilution factor corrected values were plotted (mean +/- SD, n=2). The mean NF-L concentration was determined to be 643,742 pg/mL in mouse brain and 127,718 pg/mL in rat brain tissue extract. There was no detectable signal in U-87 MG cell extract.

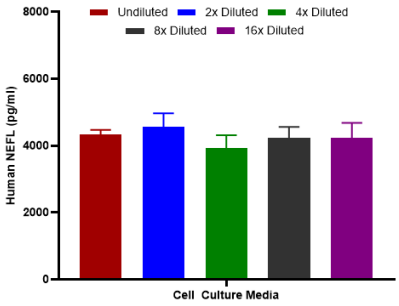

Fig3: Interpolated concentrations of spiked NF-L in cell culture media samples.

The concentrations of NF-L were measured in duplicates, interpolated from the NF-L standard curves and corrected for sample dilution. Undiluted samples are as follows: cell culture media 50%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2).

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