Encor Biotechnology Inc. NF-L DegenoTag[™] Peptide Mouse Monoclonal Antibody

RPCA-NF-L-Degen

Species Cross-Reactivity

Ordering Information Web www.encorbio.com Email admin@encorbio.com Phone 352-372-7022 Fax 352-372-7066

HGNC Name: NEFL UniProt: P07196 RRID: AB 2923499 Immunogen: Proprietary recombinant constrict containing amino acids of human NF-L expressed in and purified from E. coli.

Format: Purified antibody at 1mg/mL in 50% PBS, 50% glycerol plus 5mM NaN, Storage: Shipped on ice. Store at 4°C for short term, for longer term at -20°C. Avoid freeze / thaw cycles.

Recommended dilutions: WB: 1:1,000-1:2,000. ICC/IF: 1:10,000 IHC: 1:5,000

References:

1. Hoffman et al. Neurofilament gene expression:a major determinant of axonal caliber. PNAS 84:3472-6 (1987). 2. Perrot R, et al. Review of the Multiple Aspects

of Neurofilament Functions, and their Possible Contribution to Neurodegeneration. Mol. Neurobiol. 38:27-65 (2008). 3. Lépinoux-Chambaud C. Eyer J. Review on

intermediate filaments of the nervous system and their pathological alterations. Histochem. Cell Biol. 140:13-22 (2013).

4. Liu Q. et al. Neurofilamentopathy in Neurodegenerative Diseases. Open Neurol. J. 5:58-62 (2011).

5. Bacioglu M, et al. Neurofilament light chain in blood and CSF as marker of disease progression in mouse models and in neurodegenerative diseases. Neuron 91:56-66 (2016).

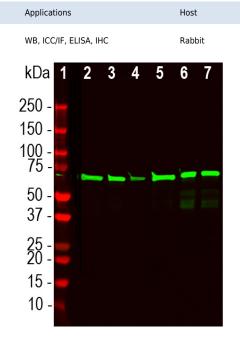
6. Shaw G, et al. Uman Type NF-L Antibodies Are Effective Reagents for the Imaging of Neurodegeneration. BioRxiv DOI

10.1101/2022.08.27.504533 (2022)

7. Norgren N, et al. Monoclonal antibodies selective for low molecular weight neurofilaments. Hybrid Hybridomics 21:53-59 (2002)

(2002). 8. Shaw G, et al. Uman type neurofilament light antibodies are effective reagents for the imaging of neurodegeneration. Brain Communications

doi.org/10.1093/braincomms/fcad067.



Western blot analysis of different tissue lysates using rabbit pAb to degenerated forms of NF-L, RPCA-NF-L-Degen, dilution 1:2,000 in green: [1] protein standard, [2] rat brain, [3] rat spinal cord, [4] mouse brain, [5] mouse spinal cord, [6] cow spinal cord and [7] pig spinal cord. The strong band at about 68kDa corresponds to full length denatured NF-L protein.

68-70kDa by SDS-PAGE Hu, Rt, Ms, Co, Pi

Molecular Wt.

Immunofluorescence of a section of spinal cord from a rat given a C4 contusion injury 3 days previously. The section was stained with mouse monoclonal to NF-M MCA-3H11 in green and counterstained with RPCA-NF-L-Degen at 1:1,000 in red. The RPCA-NF-L-Degen antibody does not stain the undamaged axons which are strongly positive for the NF-M antibody. However linear arrays of swollen profiles which originated from damaged axons are strongly positive for the rabbit NF-L-Degen antibody but not the NF-M antibody, although there is clearly some staining. The MCA-3H11 epitope, which is in the C-terminal "tail" of NF-M, has either been partially removed or destroyed.

Background:

Neurofilaments are major components of neurons and their axons (1-5). We have recently developed a series of novel reagents which we call DegenoTag[™] products. These are antibodies which recognize epitopes in a small segment of the neurofilament NF-L subunit which are normally not accessible to antibodies but which became available on degeneration (6). We propose that these epitopes are made accessible as a result of degeneration induced proteolysis, and in agreement with this hypothesis we could make previously negative control tissues become strongly DegenoTag[™] antibody positive by treatment with proteases. In addition fresh CNS tissues did not stain with DegenoTag[™] reagents except for a tiny minority of apparently spontaneously degenerating processes. In stark contrast tissues left to sit at room temperature for 4 hours were strongly reactive with DegenoTag[™] reagents. We also discovered that our antibodies to the C-terminal of NF-L, such as our rabbit polyclonal RPCA-NF-L-ct and mouse monoclonal MCA-DA2. Our reagents can therefore be used to positively identify both healthy and degenerated processes.

Isotype

RPCA-NF-L-Degen was raised against a proprietary recombinant immunogen containing amino acids 311-375 of the human NF-L sequence (6,8). The antibody works well on western blots of a variety of species but binds only degenerated processes in sectioned material. It also works well on paraffin embedded histological sections of human brain and is an excellent capture reagent in ELISA. This and other DegenoTag[™] reagents can be used to identify degenerating and degenerated processes and also to monitor NF-L degradation in a variety of contexts. Full details of these findings are described in our a BioRxiv and in greater detail in a peer-reviewed publication in Brain Communications. It also works well on paraffin embedded histological sections of rodent CNS tissues, including transgenic mouse models. EnCor also markets other DegenpoTag[™] reagents such as MCA-1D44, MCA-6H63 and MCA-1B11, three mouse monoclonal antibodies each with different epitopes on NF-L. We also market a chicken polyclonal with similar specificity e than MCA-6H63 and the rabbit polyclonal CPCA-NF-L-Degen.

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Abbreviation Key:

mAb—Monoclonal Antibody pAb—Polyclonal Antibody WB—Western Blot IF—Immunofluorescence ICC—Immunocytochemistry IHC—Immunohistochemistry E—ELISA Hu—Human Mo—Monkey Do—Dog Rt—Rat Ms—Mouse Co—Cow Pi—Pig Ho—Horse Ch—Chicken Dr—D. rerio Dm—D. melanogaster Sm—S. mutans Ce—C. elegans Sc—S. cerevisiae Sa—S. aureus Ec—E. coli.