Degenotag™ NF-L peptide Chicken Polyclonal Antibody

CPCA-NF-L-Degen

Ordering Information
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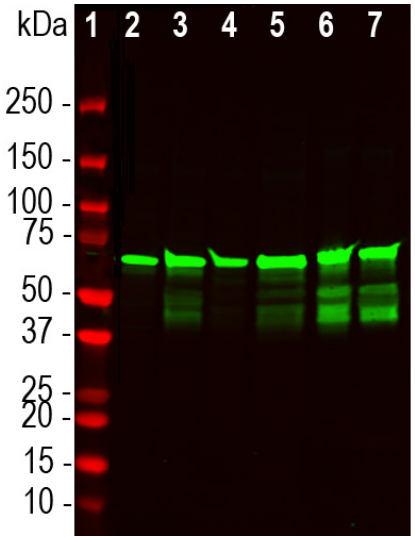
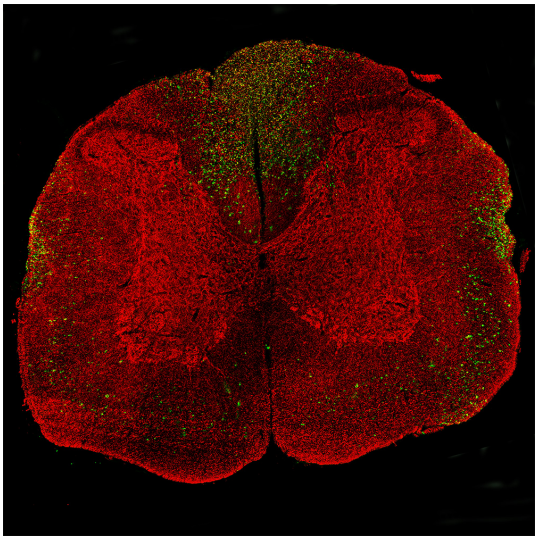
HGNC Name: *NEFL*
UniProt: *P07196*
RRID: *AB_2923500*
Immunogen: *Degenotag™ NF-L peptide*
Format: *Purified antibody at 1mg/mL in 50% PBS, 50% glycerol plus 5mM azide*
Storage: *Stable at 4°C for one year, for longer term store at -20°C*
Recommended dilutions:
WB: 1:20,000 IF/ICC: 1:10,000 IHC: 1:5,000

Applications	Host	Isotype	Molecular Wt.	Species Cross-Reactivity
WB, IF/ICC, IHC	Chicken		68-70kDa	Hu, Rt, Ms, Pi, Co



Western blot analysis of different tissue lysates using chicken pAb degenerated forms of NF-L, CPCA-NF-L-Degen dilution 1:10,000 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. Lower molecular weight bands detected mainly in the spinal cord samples are proteolytic forms of NF-L.



Immunostaining of a coronal section of the spinal cord of a rat given a midline C4 contusion injury three days previously. Sections were stained with RPCA-NF-L-ct (red) and CPCA-NF-L-Degen in green. CPCA-NF-L-Degen stains prominent aggregates of material concentrated in the lateral funiculi and the dorsal columns but seen in lesser amounts throughout the section. These are degenerating and degenerated axons damaged by the C4 lesion. The RPCA-NF-L-ct antibody binds the C-terminal "tail" region of NF-L which is absent or destroyed during degeneration, so the CPCA-NF-L-Degen positive profiles are largely negative for RPCA-NF-L-ct.

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. 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.

CPCA-NF-L-Degen was raised against a proprietary recombinant immunogen containing amino acids 311-375 of the human NF-L sequence. The antibody works well on western blots of a variety of species but binds only degenerated processes in sectioned material. We recommend using the antibody at 1:10,000 dilution for the specific visualization of degenerating and degenerated processes by IF/ICC. Full details of these findings are described in our a BioRχiv 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. 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. 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 will also market a rabbit polyclonal with similar specificity RPCA-NF-L-Degen.

References:

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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. BioRχiv 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).

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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.