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Immunogen: Recombinant construct containing the C-terminus of the human sequence (amino acids 708-877) expressed in and purified from E. coli.

Format: Concentrated IgY preparation in PBS plus

Storage: Store at 4°C for short term, for longer term

store at -20°C

Recommended dilutions:

WB: 1:2,000-5,000, IF/ICC & IHC: 1:2,000

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 O. 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. The use and potential of pNF-H as a general blood biomarker of axonal loss: an immediate application for CNS injury. in Brain Neurotrauma: Molecular, Neuropsychological, and Rehabilitation Aspects. CRC Press/Taylor & Francis Chapter 21 (2015). 7. Harris J, Ayyub C. and Shaw G. A molecular dissection of the carboxyterminal tails of the major neurofilament subunits NF-M and NF-H. J Neurosci. Res. 30:47-62 (1991). 8. Shaw G. Identification of previously unrecognized sequence motifs at the extreme carboxyterminus of the neurofilament subunit NF-M. BBRC 14;162:294-9 (1989).

Neurofilament NF-M Chicken Polyclonal Antibody

Host

Isotype

CPCA-NF-M

Species Cross-Reactivity

Hu, Rt, Ms, Bo, Po, Ho, Ck

WB, IF/ICC, IHC					Chicken						
kDa	1	2	3	4	5	6	7	8	9	10	
250-	-										
150-		1									
100-											
75-											
50-)	

Molecular Wt.

PAGE

145-160kDa by SDS-

Western blot analysis of different neuronal tissue and cell lysates using chicken pAb to NF-M, CPCA-NF-M, dilution 1:2,000 in green: [1] protein standard (red), [2] rat brain [3] rat spinal cord, [4] mouse brain, [5] mouse spinal cord, [6] NIH/3T3 cells, [7] HEK293, [8] HeLa, [9] SH-SY5Y, and [10] C6 cells. Strong band at 145kDa corresponds to rodent NF-M, and about 160kDa band corresponds to human NF-M protein, visible in SHSY-SY and HEK293 cells which have neuronal properties. NF-M is not expressed in HeLa and other cell lines tested.

Immunofluorescent analysis of rat cerebellum section stained with chicken pAb to NF-M, CPCA-NF-M, dilution 1:1,000 in red, and costained with mouse mAb to CNP, MCA-1H10, dilution 1:500 in green. The blue is DAPI staining of nuclear DNA. Following transcardial perfusion of rat with 4% paraformaldehyde, brain was post fixed for 24 hours, cut to $45\mu\text{M}$, and free-floating sections were stained with the above antibodies. The NF-M antibody labels the network of axons of basket neurons and other neurons. The CNP antibody stains oligodendrocytes, cells that create myelin sheaths around axons

Background:

Applications

Neurofilaments are the 10nm or intermediate filament proteins found specifically in neurons, and are composed predominantly of three major proteins called NF-L, NF-M and NF-H. NF-M is the neurofilament middle or medium molecular weight polypeptide and runs on SDS-PAGE gels at 145-160kDa, with some species variability, though the real molecular weight is ~105kDa. The major function of neurofilaments is likely to control the diameter of large axons (1). Antibodies to NF-M such as CPCA-NF-M are useful for identifying neuronal cells and their processes in tissue sections and in cell culture. NF-M antibodies can also be useful to visualize neurofilament rich accumulations seen in many neurological diseases, such as Amyotrophic Lateral Sclerosis (a.k.a. Lou Gehrig's disease) and Alzheimer's disease (2-4). Much recent evidence has suggested that the detection of NF-L and NF-H in blood and CSF might be a useful prognostic or diagnostic biomarkers of neuronal damage and degeneration associated with a variety of CNS pathologies (5,6). The potential utility of NF-M in this fashion has not to date been examined. The CPCA-NF-M antibody was made against a recombinant fusion protein of *E. coli* TrpE fused to the C-terminus of rat NF-M, amino acids 677-845 (7). This region is very highly conserved in protein sequence across species boundaries and contains some interesting peptide repeats of currently unknown function (8). The CPCA-NF-M antibody is very similar in properties to a rabbit polyclonal the production and characterization of which were described in reference 7. Also available from EnCor is a rabbit polyclonal and a widely used mouse monoclonal antibody to the same immunogen RPCA-NF-M, and MCA-3H11. All three antibodies works on a variety of species and are clean and specific on western blots, cell and tissue staining.

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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.