Anti-NEFM Antibody [JM11-20]

ET1703-57



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP
Molecular Wt:	Predicted band size: 102 kDa
Clone number:	JM11-20
Description:	Neurofilament-M (NF-M), for neurofilament medium polypeptide, a member of the intermediate filament family, is a major component of neuronal cytoskeletons. Neurofilaments are dynamic structures; they contain phosphorylation sites for a large number of protein kinases, including protein kinase A, protein kinase C, cyclin-dependent kinase 5, extracellular signal regulated kinase, glycogen synthase kinase-3, and stress-activated protein kinase gamma. In addition to their role in the control of axon caliber, neurofilaments may affect other cytoskeletal elements, such as microtubules and actin filaments. Changes in neurofilament phosphorylation or metabolism are frequently observed in neurodegenerative diseases, including amyotrophic lateral sclerosis (ALS), Parkinson's disease, and Alzheimer's disease.
Immunogen:	Synthetic peptide within Human NEFM aa 651-692 / 916.
Positive control:	293T cell lysates, N2A, PC-12, SH-SY5Y, mouse brain tissue, rat brain tissue, human brain tissue.
Subcellular location:	Cell projection, Cytoplasm, Cytoskeleton, Intermediate filament.
Database links:	SwissProt: P07197 Human P08553 Mouse P12839 Rat
Recommended Dilutions: WB IF-Cell IF-Tissue IHC-P IP	1:1,000-1:2,000 1:100-1:500 1:100-1:500 1:50-1:200 Use at an assay dependent concentration.
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!\!C$ after thawing. Aliquot store at -20 $^\circ\!\!C$ or -80 $^\circ\!\!C$. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

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Orders:0086-571-88062880

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Images

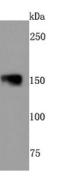


Fig1: Western blot analysis of NEFM on 293T cell lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET1703-57, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

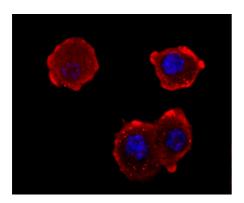


Fig2: ICC staining of NEFM in N2A cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1703-57, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

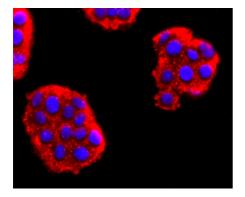


Fig3: ICC staining of NEFM in PC-12 cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1703-57, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

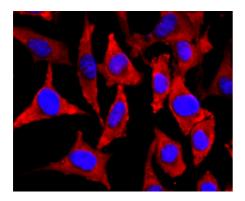


Fig4: ICC staining of NEFM in SH-SY5Y cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1703-57, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

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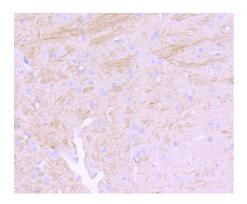


Fig5: Immunohistochemical analysis of paraffin-embedded mouse brain tissue using anti-NEFM antibody. The section was pretreated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1703-57, 1/50) for 30 minutes 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.

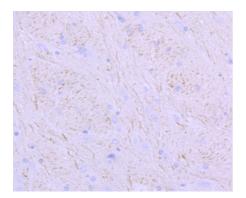


Fig6: Immunohistochemical analysis of paraffin-embedded rat brain tissue using anti-NEFM antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1703-57, 1/50) for 30 minutes 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.



Fig7: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-NEFM antibody (ET1703-57) at 1/400 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 ddH₂O and PBS, and then probed with the primary antibody (ET1703-57) at 1/400 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.

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Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

- 1. Guven S et al. Functional maintenance of differentiated embryoid bodies in microfluidic systems: a platform for personalized medicine. Stem Cells Transl Med 4:261-8 (2015).
- 2. Khan RS et al. SIRT1 Activating compounds reduce oxidative stress mediated neuronal loss in viral induced CNS demyelinating disease. Acta Neuropathol Commun 2:3 (2014).

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