abcam

Product datasheet

Anti-Neurofilament heavy polypeptide antibody [3G3] ab19386

5 References 4 Images

Overview

Product name	Anti-Neurofilament heavy polypeptide antibody [3G3]
Description	Mouse monoclonal [3G3] to Neurofilament heavy polypeptide
Host species	Mouse
Tested applications	**Suitable for:** ICC/IF, IHC-P, WB
Species reactivity	**Reacts with:** Rat, Human
Immunogen	Synthetic peptide corresponding to Neurofilament heavy polypeptide aa 1-226 (C terminal).
Positive control	ICC: Rat brain microsome. WB: SH-SY5Y cells, rat brain lysates. IHC-P: Rat and human brain tissue.
General notes	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer	Preservative: 0.05% Sodium azide Constituents: PBS, 0.1% BSA
Purity	Protein G purified
Clonality	Monoclonal
Clone number	3G3
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab19386 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF		Use a concentration of 5 µg/ml.
IHC-P		1/10 - 1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		1/100 - 1/1000. Predicted molecular weight: 112 kDa.

Target

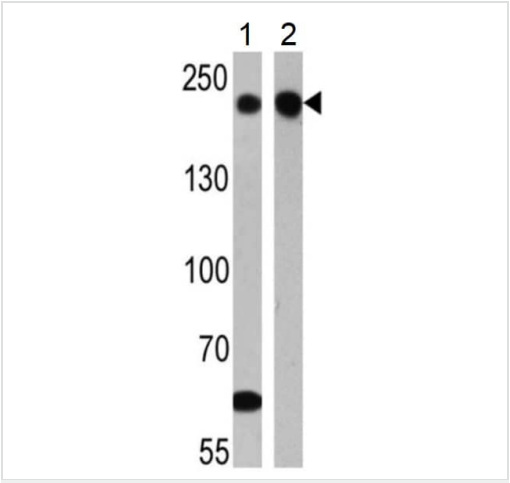
Function Neurofilaments usually contain three intermediate filament proteins: L, M, and H which are involved in the maintenance of neuronal caliber. NF-H has an important function in mature axons that is not subserved by the two smaller NF proteins.

Involvement in disease Defects in NEFH are a cause of susceptibility to amyotrophic lateral sclerosis (ALS) [MIM:105400]. ALS is a neurodegenerative disorder affecting upper and lower motor neurons, and resulting in fatal paralysis. Sensory abnormalities are absent. Death usually occurs within 2 to 5 years. The etiology is likely to be multifactorial, involving both genetic and environmental factors.

Sequence similarities Belongs to the intermediate filament family.

Post-translational modifications There are a number of repeats of the tripeptide K-S-P, NFH is phosphorylated on a number of the serines in this motif. It is thought that phosphorylation of NFH results in the formation of interfilament cross bridges that are important in the maintenance of axonal caliber.
Phosphorylation seems to play a major role in the functioning of the larger neurofilament polypeptides (NF-M and NF-H), the levels of phosphorylation being altered developmentally and coincident with a change in the neurofilament function.
Phosphorylated in the Head and Rod regions by the PKC kinase PKN1, leading to inhibit polymerization.

Images



Western blot - Anti-Neurofilament heavy polypeptide antibody [3G3] (ab19386)

All lanes : Anti-Neurofilament heavy polypeptide antibody [3G3] (ab19386) at 1/500 dilution

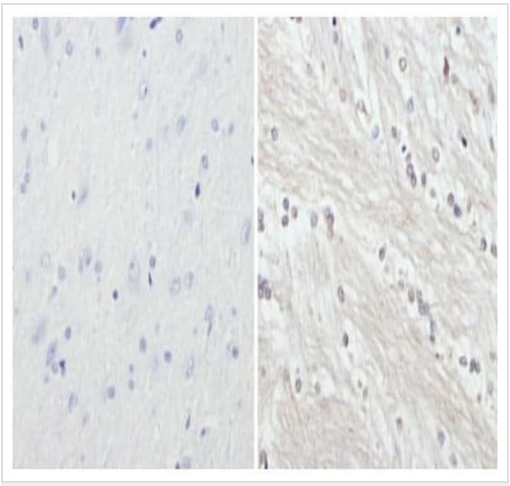
Lane 1 : SH-SY5Y (human neuroblastoma cell line from bone marrow) whole cell lysate
Lane 2 : Rat brain tissue lysates

Lysates/proteins at 25 µg per lane.

Secondary
All lanes : HRP-conjugated secondary antibody

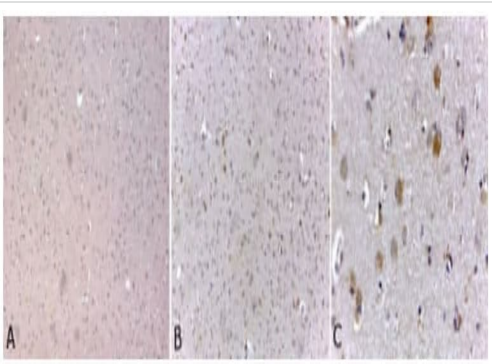
Predicted band size: 112 kDa

Results show a band at ~200 kDa.



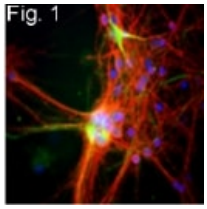
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neurofilament heavy polypeptide antibody [3G3] (ab19386)

Immunohistochemistry analysis of paraffin-embedded rat brain tissue (right) compared to a negative control without primary antibody (left) labelling Neurofilament Heavy Chain with ab19386. Positive staining in the cytoplasm and nucleus (right).
To expose target proteins, antigen retrieval was performed using 10mM sodium citrate (pH 6.0), microwaved for 8-15 min. Following antigen retrieval, tissues were blocked in 3% H2O2-methanol for 15 min at room temperature, washed with ddH2O and PBS, and then probed with a Neurofilament Heavy Chain monoclonal antibody diluted in 3% BSA-PBS at 1/20 dilution overnight at 4°C in a humidified chamber. Tissues were washed extensively in PBST and detection was performed using an HRP-conjugated secondary antibody followed by colorimetric detection using a DAB kit. Tissues were counterstained with hematoxylin and dehydrated with ethanol and xylene to prep for mounting.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Neurofilament heavy polypeptide antibody [3G3] (ab19386)

Immunohistochemistry analysis of Neurofilament, Heavy chain showing staining in the cytoplasm and axons of formalin-fixed, paraffin-embedded human brain tissue (B) and magnified section (C) compared with an isotype control (A). To expose target proteins, antigen retrieval was performed using HEIR with a buffer (pH 6.2). Tissues were probed with a Neurofilament, Heavy chain monoclonal antibody for 60 minutes at a dilution of 2 µg/mL and detection was performed using an HRP-conjugated detection system for 30 minutes followed by DAB staining.



Immunocytochemistry/ Immunofluorescence - Anti-Neurofilament heavy polypeptide antibody [3G3] (ab19386)

Immunofluorescence of neurofilament, heavy chain in rat cerebral cortex cultures in green.

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