Human/Mouse/Rat NF-L Antibody

Monoclonal Mouse IgG_{2B} Clone # 268014

Catalog Number: MAB2216

DESCRIPTION

Species Reactivity	Human/Mouse/Rat
Specificity	Detects human NF-L in direct ELISAs and human, mouse and rat NF-L in Western blots. In direct ELISAs and Western blots, no cross-reactivity with recombinant human (rh) NF-H or rhNF-M is observed.
Source	Monoclonal Mouse IgG_{2B} Clone # 268014
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	*E. coli*-derived recombinant human NF-L Met1-Asp338 Accession # P07196
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.

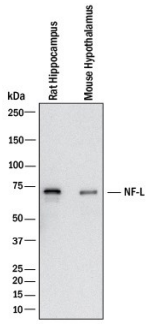
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	1 µg/mL	Mouse brain (hypothalamus) and rat brain (hippocampus)
Immunocytochemistry	8 µg/mL	Immersion fixed U251 human myeloma cell line (positive) and K562 human chronic myelogenous leukemia cell line (negative)
Immunohistochemistry	3-25 µg/mL	See Below

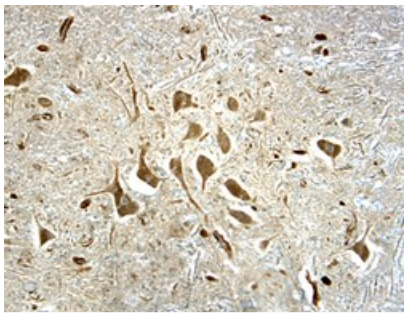
DATA

Western Blot



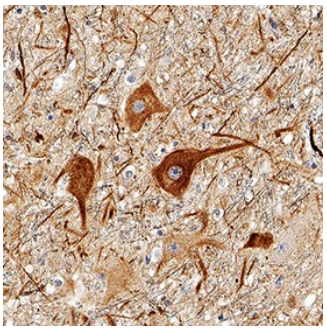
Detection of Mouse and Rat NF-L by Western Blot. Western blot shows lysates of mouse brain (hypothalamus) and rat brain (hippocampus). PVDF membrane was probed with 1 µg/mL of Mouse Anti-Human/Mouse/Rat NF-L Monoclonal Antibody (Catalog # MAB2216) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018) . A specific band was detected for NF-L at approximately 74 kDa (as indicated). This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Immunohistochemistry



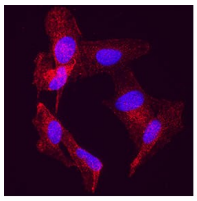
NF-L in Human Spinal Cord. NF-L was detected in immersion fixed paraffin-embedded sections of human spinal cord using 25 µg/mL Mouse Anti-Human NF-L Monoclonal Antibody (Catalog # MAB2216) overnight at 4 °C. Tissue was stained with the Anti-Mouse HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS002) and counter-stained with hematoxylin (blue). Specific labeling was localized to the cytoplasm in motor neurons. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Immunohistochemistry

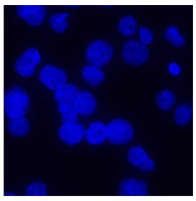


NF-L in Human Brain. NF-L was detected in immersion fixed paraffin-embedded sections of human brain (substantia nigra) using Mouse Anti-Human NF-L Monoclonal Antibody (Catalog # MAB2216) at 3 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC001). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to cytoplasm in neurons. View our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

Immunocytochemistry



U-251 cells (positive)



K-562 cells (negative)

NF-L in U251 human myeloma cell line (positive) and K562 human chronic myelogenous leukemia cell line (negative). NF-L was detected in immersion fixed U251 human myeloma cell line (positive) and K562 human chronic myelogenous leukemia cell line (negative) using Mouse Anti-Human/Mouse/Rat NF-L Monoclonal Antibody (Catalog # MAB2216) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.

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Monoclonal Mouse IgG_{2B} Clone # 268014

Catalog Number: MAB2216

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

NF-L is a 68 kDa light chain cytoskeletal intermediate filament protein that is expressed in neurons. It associates with the 125 kDa NF-M and the 200 kDa NF-H to form neurofilaments.

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