BOSTER BIOLOGICAL TECHNOLOGY
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# Anti-NF-M/NEFM Antibody Picoband®

**Catalog Number:** A06821-2

## About NEFM

Neurofilaments are type IV intermediate filament heteropolymers composed of light, medium, and heavy chains. Neurofilaments comprise the axoskeleton and functionally maintain neuronal caliber. They may also play a role in intracellular transport to axons and dendrites. This gene encodes the medium neurofilament protein. This protein is commonly used as a biomarker of neuronal damage. Alternative splicing results in multiple transcript variants encoding distinct isoforms.

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## Overview

| | |
|---|---|
| Product Name | Anti-NF-M/NEFM Antibody Picoband® |
| Reactive Species | Human, Mouse, Rat |
| Description | Boster Bio Anti-NF-M/NEFM Antibody Picoband® catalog # A06821-2. Tested in WB, IHC, IF, IP, Flow Cytometry, ELISA applications. This antibody reacts with Human, Mouse, Rat. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance. |
| Application | ELISA, Flow Cytometry, IP, IF, IHC, WB |
| Clonality | Polyclonal |
| Formulation | Each vial contains 4 mg Trehalose, 0.9 mg NaCl, 0.2 mg Na2HPO4. |
| Storage Instructions | At -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freezing and thawing. |
| Host | Rabbit |
| Uniprot ID | P07197 |

## Technical Details

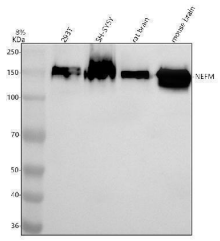
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| Immunogen | E.coli-derived human NF-M/NEFM recombinant protein (Position: Q125-D916). Human NF-M/NEFMshares 72.3% and 72% amino acid (aa) sequence identity with mouse and rat NF-M/NEFM, respectively. |

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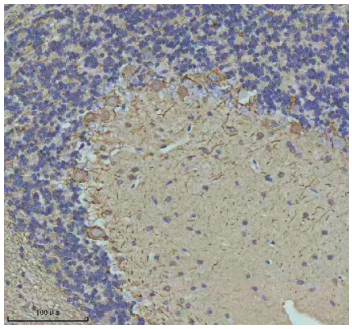
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| Form | Lyophilized |
| Concentration | Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml. |
| Purification | Immunogen affinity purified. |
| Suggested Dilutions | Western blot, 0.25-0.5 ug/ml, Human, Mouse, Rat<br>Immunohistochemistry(Paraffin-embedded Section), 2-5 ug/ml, Mouse, Rat<br>Immunoprecipitation, 0.5-2 ug/ml, Human<br>Immunofluorescence, 5 ug/ml, Mouse, Rat<br>Flow Cytometry (Fixed), 1-3 ug/1x$10^6$ cells, Human<br>ELISA, 0.1-0.5 ug/ml<br><br>Note: For flow cytometry (FCM), suitable for fixed cells only. Not suitable for FACS or surface flow cytometry using live cells. |

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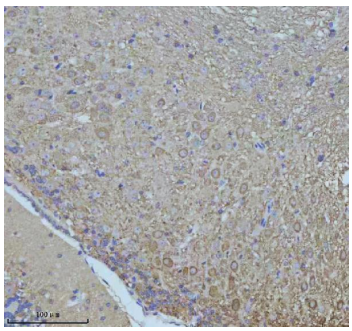
# Anti-NF-M/NEFM Antibody Picoband® (A06821-2) Images




Western blot analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). Electrophoresis was performed on a 8% SDS-PAGE gel at 80V (Stacking gel) / 120V (Resolving gel) for 2 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human 293T whole cell lysates, Lane 2: human SH-SY5Y whole cell lysates, Lane 3: rat brain tissue lysates, Lane 4: mouse brain tissue lysates.After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-NF-M/NEFM antigen affinity purified polyclonal antibody (A06821-2) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an ECL Plus Western Blotting Substrate (Catalog # AR1196-200) with Tanon 5200 system. A specific band was detected for NF-M/NEFM at approximately 160 kDa. The expected band size for NF-M/NEFM is at 102 kDa.



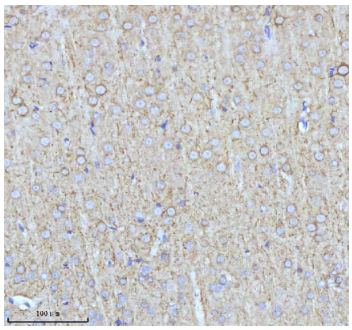
IHC analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). NF-M/NEFM was detected in a paraffin-embedded section of rat cerebellum tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-NF-M/NEFM Antibody (A06821-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



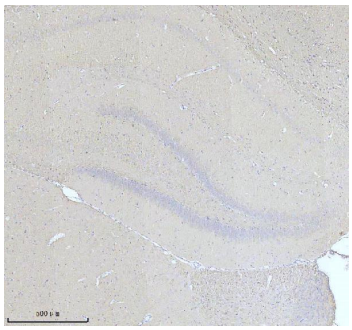
IHC analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). NF-M/NEFM was detected in a paraffin-embedded section of mouse cerebellum tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-NF-M/NEFM Antibody (A06821-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

IHC analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). NF-M/NEFM was detected in a paraffin-embedded section of rat brain tissue. Heat mediated antigen

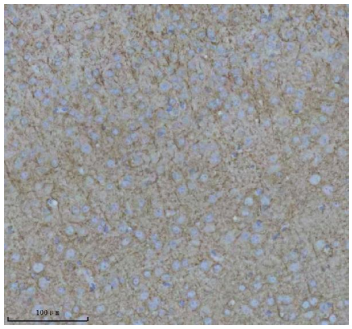
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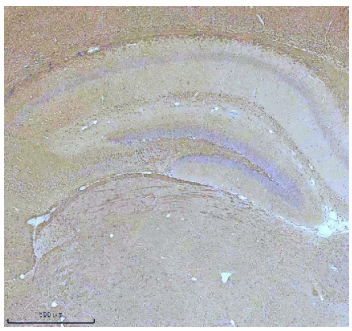
retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-NF-M/NEFM Antibody (A06821-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



IHC analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). NF-M/NEFM was detected in a paraffin-embedded section of rat brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-NF-M/NEFM Antibody (A06821-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



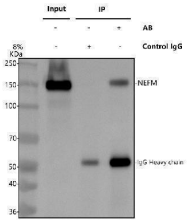
IHC analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). NF-M/NEFM was detected in a paraffin-embedded section of mouse brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-NF-M/NEFM Antibody (A06821-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



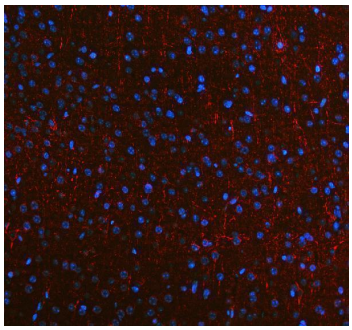
IHC analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). NF-M/NEFM was detected in a paraffin-embedded section of mouse brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-NF-M/NEFM Antibody (A06821-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

Immunoprecipitating NF-M/NEFM in SH-SY5Y whole cell lysate. Western blot analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). Lane 1: SH-SY5Y whole cell lysates (30ug), Lane 2: Rabbit control IgG instead of anti-NF-M/NEFM antibody in SH-SY5Y whole cell lysate, Lane 3: anti-

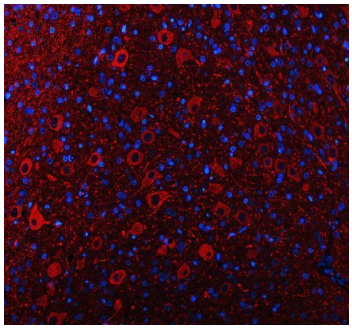
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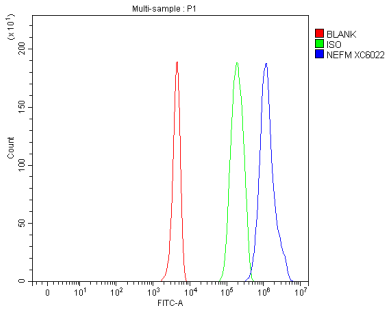

NF-M/NEFM antibody (2ug) + SH-SY5Y whole cell lysate (500ug). After electrophoresis, proteins were transferred to a membrane. Then the membrane was incubated with rabbit anti-NF-M/NEFM antigen affinity purified polyclonal antibody (A06821-2) at a dilution of 0.5 ug/mL and probed with a goat anti-rabbit IgG-HRP secondary antibody (Catalog # BA1054). The signal is developed using ECL Plus Western Blotting Substrate (Catalog # AR1197). A specific band was detected for NF-M/NEFM at approximately 160 kDa. The expected band size for NF-M/NEFM is at 102 kDa.



IF analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). NF-M/NEFM was detected in a paraffin-embedded section of mouse brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 ug/mL rabbit anti-NF-M/NEFM Antibody (A06821-2) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) was used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



IF analysis of NF-M/NEFM using anti-NF-M/NEFM antibody (A06821-2). NF-M/NEFM was detected in a paraffin-embedded section of rat cerebellum tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 ug/mL rabbit anti-NF-M/NEFM Antibody (A06821-2) overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG (BA1032) was used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.




Flow Cytometry analysis of 293T cells using anti-NF-M/NEFM antibody (A06821-2). Overlay histogram showing 293T cells stained with A06821-2 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-NF-M/NEFM Antibody (A06821-2, 1 ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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# Anti-NF-M/NEFM Antibody

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