Product datasheet

Anti-NF2 / Merlin antibody [AF1G4] ab88957

Overview

Product name Anti-NF2 / Merlin antibody [AF1G4]
Description Mouse monoclonal [AF1G4] to NF2 / Merlin
Host species Mouse
Tested applications Suitable for: ICC/IF, ChIP, WB, Indirect ELISA, IHC-P
Species reactivity Reacts with: Mouse, Rat, Human
Immunogen Recombinant human NF2/ Merlin with His tag purified from E.coli
Positive control HeLa, 293T, MCF7, HepG2, MOLT4, C6 and L929 cell lysates. Human skeletal muscle FFPE tissue sections
General notes This product was changed from ascites to tissue culture supernatant on 18th September 2017. Lot number GR310755 and higher lots are from tissue culture supernatant. Please note that the dilutions may need to be adjusted accordingly.

Properties

Form Liquid
Storage instructions Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid repeated freeze / thaw cycles.
Storage buffer Preservative: 0.03% Sodium azide
Constituents: HEPES, 0.01% BSA, 50% Glycerol, 0.87% Sodium chloride
Purity Tissue culture supernatant
Clonality Monoclonal
Clone number AF1G4
Isotype IgG2a
Light chain type kappa

Applications

Our Abpromise guarantee covers the use of ab88957 in the following tested applications.
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.
**Function**
Probable regulator of the Hippo/SWH (Sav/Wts/Hpo) signaling pathway, a signaling pathway that plays a pivotal role in tumor suppression by restricting proliferation and promoting apoptosis. Along with WWC1 can synergistically induce the phosphorylation of LATS1 and LATS2 and can probably function in the regulation of the Hippo/SWH (Sav/Wts/Hpo) signaling pathway. May act as a membrane stabilizing protein. May inhibit PI3 kinase by binding to AGAP2 and impairing its stimulating activity. Suppresses cell proliferation and tumorigenesis by inhibiting the CUL4A-RBX1-DDB1-VprBP/DCAF1 E3 ubiquitin-protein ligase complex.

**Tissue specificity**
Widely expressed. Isoform 1 and isoform 3 are predominant. Isoform 4, isoform 5 and isoform 6 are expressed moderately. Isoform 8 is found at low frequency. Isoform 7, isoform 9 and isoform 10 are not expressed in adult tissues, with the exception of adult retina expressing isoform 10. Isoform 9 is faintly expressed in fetal brain, heart, lung, skeletal muscle and spleen. Fetal thymus expresses isoforms 1, 7, 9 and 10 at similar levels.

**Involvement in disease**
Defects in NF2 are the cause of neurofibromatosis 2 (NF2) [MIM:101000]; also known as central neurofibromatosis. NF2 is a genetic disorder characterized by bilateral vestibular schwannomas (formerly called acoustic neuromas), schwannomas of other cranial and peripheral nerves, meningiomas, and ependymomas. It is inherited in an autosomal dominant fashion with full penetrance. Affected individuals generally develop symptoms of eighth-nerve dysfunction in early adulthood, including deafness and balance disorder. Although the tumors of NF2 are histologically benign, their anatomic location makes management difficult, and patients suffer great morbidity and mortality. 
Defects in NF2 are a cause of schwannomatosis (SCHWA) [MIM:162091]; also known as congenital cutaneous neurilemmomatosis. Schwannomas are benign tumors of the peripheral nerve sheath that usually occur singly in otherwise normal individuals. Multiple schwannomas in the same individual suggest an underlying tumor-predisposition syndrome. The most common such syndrome is NF2. The hallmark of NF2 is the development of bilateral vestibular-nerve schwannomas; but two-thirds or more of all NF2-affected individuals develop schwannomas in other locations, and dermal schwannomas may precede vestibular tumors in NF2-affected children. There have been several reports of individuals with multiple schwannomas who do not show evidence of vestibular schwannoma. Clinical report suggests that schwannomatosis is a clinical entity distinct from other forms of neurofibromatosis.

**Sequence similarities**
Contains 1 FERM domain.

**Post-translational**
Phosphorylation of Ser-518 inhibits nuclear localization by disrupting the intramolecular interactions of the protein.
modifications

Ubiquitinated by the CUL4A-RBX1-DDB1-DCAF1/VprBP E3 ubiquitin-protein ligase complex for ubiquitination and subsequent proteasome-dependent degradation.

Cellular localization

Cytoplasm > perinuclear region. Cytoplasmic granule. Observed in cytoplasmic granules concentrated in a perinuclear location. Isoform 7 is absent from ruffling membranes and filopodia; Cytoplasm > perinuclear region. Cytoplasmic granule. Observed in cytoplasmic granules concentrated in a perinuclear location. Isoform 9 is absent from ruffling membranes and filopodia; Nucleus. Cell projection > filopodium membrane. Cell projection > ruffle membrane. Cytoplasm > perinuclear region. Cytoplasmic granule. Cytoplasm > cytoskeleton. In a fibroblastic cell line, isoform 10 is found homogeneously distributed over the entire cell, with a particularly strong staining in ruffling membranes and filopodia and Cell projection > filopodium membrane. Cell projection > ruffle membrane. Nucleus. In a fibroblastic cell line, isoform 1 is found homogeneously distributed over the entire cell, with a particularly strong staining in ruffling membranes and filopodia. Colocalizes with MPP1 in non-myelin-forming Schwann cells. Binds with VPRBP in the nucleus. The intramolecular association of the FERM domain with the C-terminal tail promotes nuclear accumulation. The unphosphorylated form accumulates predominantly in the nucleus while the phosphorylated form is largely confined to the non-nuclear fractions.

Images

Lane 1: Wild-type HAP1 cell lysate (40 µg)
Lane 2: Empty lane
Lane 3: NF2 / Merlin knockout HAP1 cell lysate (20 µg)
Lane 4: Empty lane
Lanes 1 - 4: Merged signal (red and green). Green - ab88957 observed at 70 kDa. Red - loading control, ab181602, observed at 37 kDa.

ab88957 was shown to specifically react with NF2/Merlin in wild-type HAP1 cells. No band was observed when NF2 / Merlin knockout samples were examined. Wild-type and NF2/Merlin knockout samples were subjected to SDS-PAGE. Ab88957 and ab181602 (loading control to GAPDH) were diluted at 1/500 and 1/10,000 dilution respectively and incubated overnight at 4°C. Blots were developed with IRDye® 800CW Goat anti-Mouse IgG (H + L) ab216772 and IRDye® 680 Goat anti-Rabbit IgG (H + L) ab216777 secondary antibodies at 1/10,000 dilution for 1 hour at room temperature before imaging.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NF2 / Merlin antibody [AF1G4] (ab88957)

IHC image of NF2/Merlin staining in human skeletal muscle formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab88957, 10µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

Immunocytochemistry/ Immunofluorescence - Anti-NF2 / Merlin antibody [AF1G4] (ab88957)

ICC/IF image of ab88957 stained HeLa cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab88957, 1/200 dilution) overnight at +4°C. The secondary antibody (green) was ab88957, DyLight® 488 goat anti-mouse IgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.
<table>
<thead>
<tr>
<th>Lane</th>
<th>Lysate Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>HeLa cell lysate</td>
</tr>
<tr>
<td>2</td>
<td>293T cell lysate</td>
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<tr>
<td>3</td>
<td>MCF7 cell lysate</td>
</tr>
<tr>
<td>4</td>
<td>HepG2 cell lysate</td>
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<tr>
<td>5</td>
<td>MOLT4 cell lysate</td>
</tr>
<tr>
<td>6</td>
<td>C6 cell lysate</td>
</tr>
<tr>
<td>7</td>
<td>L929 cell lysate</td>
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</tbody>
</table>

**Predicted band size**: 69 kDa

**Observed band size**: 69 kDa

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**Western blot - Anti-NF2 / Merlin antibody [AF1G4] (ab88957)**

**All lanes**: Anti-NF2 / Merlin antibody [AF1G4] (ab88957) at 1/2000 dilution

Please note: All products are “FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES”

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