abcam

Product datasheet

Anti-Neurofibromin antibody [EPR22989-68] ab238142





5 Images

Overview

Product name Anti-Neurofibromin antibody [EPR22989-68]

Description Rabbit monoclonal [EPR22989-68] to Neurofibromin

Host species Rabbit

Suitable for: WB **Tested applications**

Unsuitable for: Flow Cyt,ICC/IF,IHC-P or IP

Species reactivity Reacts with: Mouse, Rat, Human

Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. **Immunogen**

Positive control WB: HAP1, HeLa, Mouse brain, Rat brain, NIH/3T3, MEF and PC-12 whole cell lysates.

General notes This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal EPR22989-68 Clone number

Isotype ΙgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab238142 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
WB		1/1000. Predicted molecular weight: 319 kDa.

Application notes

Is unsuitable for Flow Cyt,ICC/IF,IHC-P or IP.

Target

Function

Stimulates the GTPase activity of Ras. NF1 shows greater affinity for Ras GAP, but lower specific activity. May be a regulator of Ras activity.

Involvement in disease

Defects in NF1 are the cause of neurofibromatosis type 1 (NF1) [MIM:162200]; also known as von Recklinghausen syndrome. A disease characterized by patches of skin pigmentation (cafe-au-lait spots), Lisch nodules of the iris, tumors in the peripheral nervous system and fibromatous skin tumors. Individuals with the disorder have increased susceptibility to the development of benign and malignant tumors.

Defects in NF1 are a cause of juvenile myelomonocytic leukemia (JMML) [MIM:607785]. JMML is a pediatric myelodysplastic syndrome that constitutes approximately 30% of childhood cases of myelodysplastic syndrome (MDS) and 2% of leukemia. Germline mutations of NF1 account for the association of JMML with type 1 neurofibromatosis (NF1).

Defects in NF1 are the cause of Watson syndrome (WS) [MIM:193520]. WS is characterized by the presence of pulmonary stenosis, cafe-au-lait spots, and mental retardation. WS is considered as an atypical form of NF1.

Defects in NF1 are a cause of familial spinal neurofibromatosis (FSNF) [MIM:162210]. Familial spinal NF is considered to be an alternative form of neurofibromatosis, showing multiple spinal tumors.

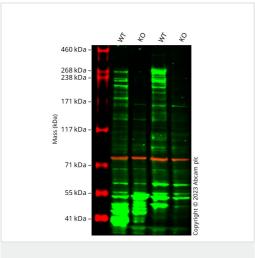
Defects in NF1 are a cause of neurofibromatosis-Noonan syndrome (NFNS) [MIM:601321]. NFNS is characterized by manifestations of both NF1 and Noonan syndrome (NS). NS is a disorder characterized by dysmorphic facial features, short stature, hypertelorism, cardiac anomalies, deafness, motor delay, and a bleeding diathesis.

Defects in NF1 may be a cause of colorectal cancer (CRC) [MIM:114500].

Sequence similarities

Contains 1 CRAL-TRIO domain. Contains 1 Ras-GAP domain.

Images



Western blot - Anti-Neurofibromin antibody [EPR22989-68] (ab238142)

All lanes : Anti-Neurofibromin antibody [EPR22989-68] (ab238142) at 1/1000 dilution

Lane 1: Wild-type MCF7 cell lysate

Lane 2: NF1 knockout MCF7 cell lysate

Lane 3 : Wild Type HeLa <u>ab255929</u> cell lysate

Lane 4 : NF1 knockout Hela <u>ab258533</u> cell lysate

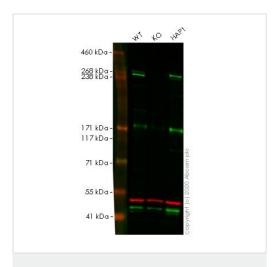
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 319 kDa

Observed band size: 180-270 kDa

Anti-NF1 antibody [EPR22989-68] (ab238142) staining at 1/1000 dilution, shown in green; Mouse anti-CANX [CANX/1543] (ab238078) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab238142 was shown to bind specifically to NF1. A band was observed at 180-270 kDa in wild-type MCF7 cell lysates with no signal observed at this size in NF1 knockout cell line. To generate this image, wild-type and NF1 knockout MCF7 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit lgG H&L 800CW and Goat anti-Mouse lgG H&L 680RD at 1/20000 dilution.



Western blot - Anti-Neurofibromin antibody [EPR22989-68] (ab238142)

All lanes : Anti-Neurofibromin antibody [EPR22989-68] (ab238142) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: NFIB knockout HeLa cell lysate

Lane 3: HAP1 cell lysate

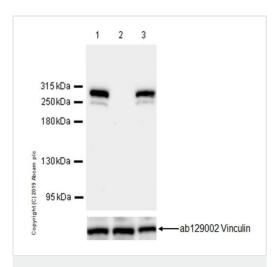
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 319 kDa

Lanes 1-3: Merged signal (red and green). Green - ab238142 observed at 319 kDa. Red - loading control <u>ab7291</u> observed at 50 kDa.

ab238142 Recombinant Anti-NF1 antibody [EPR22989-68] was shown to specifically react with NFIB in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab264725
(knockout cell lysate ab258533) was used. Wild-type and NF1 knockout samples were subjected to SDS-PAGE. ab238142 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Neurofibromin antibody [EPR22989-68] (ab238142)

All lanes : Anti-Neurofibromin antibody [EPR22989-68] (ab238142) at 1/1000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: Neurofibromin knockout HAP1 whole cell lysate

Lane 3 : HeLa (human cervix adenocarcinoma epithelial cell) whole

cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/50000 dilution

Predicted band size: 319 kDa **Observed band size:** 250,300 kDa

ab238142 was shown to specifically react with Neurofibromin in wild-type HAP1 cells as signal was lost in Neurofibromin knockout cells. Wild-type and Neurofibromin knockout samples were subjected to SDS-PAGE. ab238142 and <u>ab129002</u> (Rabbit anti-Vinculin loading control) were incubated 1 hour at room temperature at 1/1000 dilution and 1/5000 dilution respectively. Blots were developed with Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (<u>ab97051</u>) secondary antibody at 1/100,000 dilution for 1 hour at room temperature before imaging. The blot was developed on a BIO-RAD® ChemiDoc™ MP instrument using the ECL technique. Lysate should be made freshly and used in WB immediately to minimize protein degradation. Degraded fragment(250 KD) observed is consistent with what has been described in the literature (PMID: 30131853, PMID: 30408279).

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

Exposure time: 26 seconds



Western blot - Anti-Neurofibromin antibody [EPR22989-68] (ab238142)

All lanes : Anti-Neurofibromin antibody [EPR22989-68] (ab238142) at 1/1000 dilution

Lane 1: Mouse brain lysate

Lane 2: Rat brain lysate

 $\textbf{Lane 3:} \ \text{NIH/3T3 (mouse embryonic fibroblast) whole cell lysate}$

Lane 4: MEF (mouse embryonic fibroblast (immortalized)) whole

cell lysate

Lane 5: PC-12 (rat adrenal gland heochromocytoma) whole cell

lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

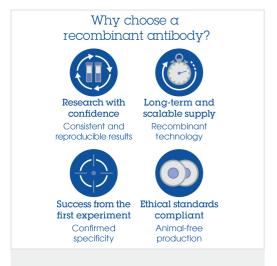
Predicted band size: 319 kDa

Observed band size: 200,250,300 kDa

Degraded fragments (200-250 KD) observed is consistent with what has been described in the literature (PMID: 30131853, PMID: 30408279).

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

Exposure time: Lane 1:3 minutes; Lane 2:26 seconds; Lanes 3-5:3 minutes.



Anti-Neurofibromin antibody [EPR22989-68] (ab238142)

Our Abpromise to you: Quality guaranteed and expert technical support

- · Replacement or refund for products not performing as stated on the datasheet
- · Valid for 12 months from date of delivery
- · Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- · We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit https://www.abcam.com/abpromise or contact our technical team.

Terms and conditions

· Guarantee only valid for products bought direct from Abcam or one of our authorized distributors