abcam

Product datasheet

Human NF2 (Merlin) knockout HeLa cell lysate ab257179

画像数 3

製品の概要

製品名	Human NF2 (Merlin) knockout HeLa cell lysate
製品の概要	Knockout cell lysate achieved by CRISPR/Cas9.
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 1 bp deletion in exon 1.
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
Reconstitution notes	To use as WB control, resuspend the lyophilizate in 50 µL of LDS* Sample Buffer to have a final concentration of 2 mg/ml. For reducing conditions, we recommend a final concentration of 0.1 M DTT. **Usage of SDS sample buffer is not recommended with these lyophilized lysates.*
特記事項	**Lysate preparation:** Our lysates are made using RIPA buffer to which we add a protease inhibitor cocktail and phosphatase inhibitor cocktail (ratio: 300:100:10). *This means that the protein of interest is denatured.* If you require a native form of the protein please use the live cell version - found **here**. Please refer to our lysis protocol for further details on how our lysates are prepared. **User storage instructions:** Lyophilizate may be stored at 4°C. After reconstitution, store at -20°C for short-term storage or -80°C for long-term storage. Access thousands of knockout cell lysates, generated from commonly used cancer cell lines. **See here for more information on knockout cell lysates.** Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of products that contain European Authorisation list (Annex XIV) substances. It is the responsibility of our customers to check the necessity of application of REACH Authorisation, and any other relevant authorisations, for their intended uses. This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our **limited use license** and **patent pages**.
アプリケーション	**適用あり:** WB

製品の特性

保存方法	Store at -80°C. Please refer to protocols.

内容	1 kit
ab260891 - Human NF2 knockout HeLa cell lysate	1 x 100µg
ab255929 - Human wild-type HeLa cell lysate	1 x 100µg

Cell type	epithelial
Disease	Adenocarcinoma
Gender	Female
STR Analysis	Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18 TH01: 7 TPOX: 8, 12 CSF1PO: 9, 10

ターゲット情報

機能	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.
組織特異性	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.
関連疾患	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.
配列類似性	Contains 1 FERM domain.

翻訳後修飾	Phosphorylation of Ser-518 inhibits nuclear localization by disrupting the intramolecular association of the FERM domain with the C-terminal tail. Ubiquitinated by the CUL4A-RBX1-DDB1-DCAF1/VprBP E3 ubiquitin-protein ligase complex for ubiquitination and subsequent proteasome-dependent degradation.
細胞内局在	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.

アプリケーション

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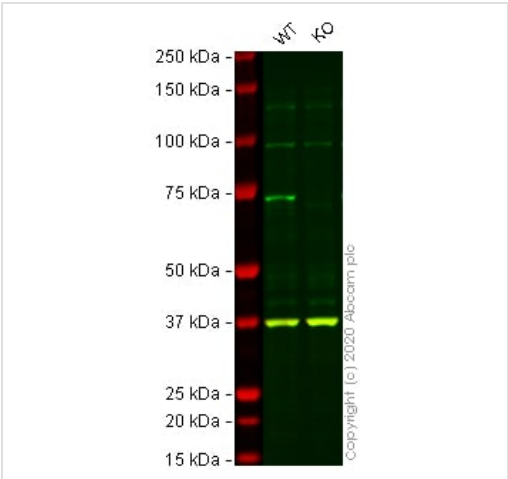
アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration. Predicted molecular weight: 70 kDa.

画像



Western blot - Human NF2 (Merlin) knockout HeLa cell lysate (ab257179)

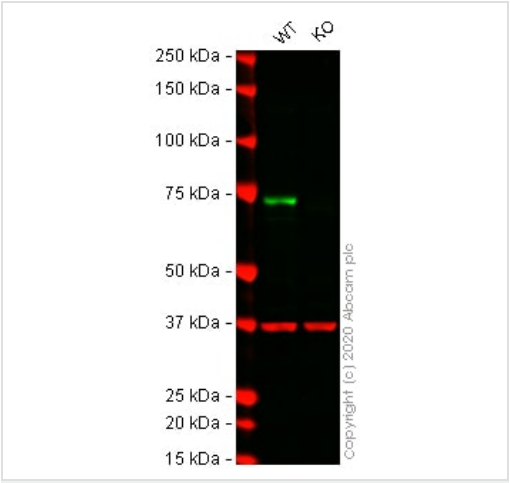
Lane 1: Wild-type HeLa cell lysate 20 ug

Lane 2: NF2 knockout HeLa cell lysate 20 ug

Lanes 1 - 2: Merged signal (red and green). Green - **ab88957** observed at 60 kDa. Red - loading control **ab181602** (Rabbit Anti-GAPDH antibody [EPR16891]) observed at 37kDa.

ab88957 was shown to react with NF2 in wild-type HeLa cells in western blot with loss of signal observed in NF2 knockout cell line **ab261796** (NF2 knockout cell lysate ab257179). Wild-type and NF2 knockout HeLa cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab88957** and **ab181602** (Rabbit Anti-GAPDH antibody [EPR16891]) overnight at 4°C at a 1 in 2000 dilution and a 1 in 20000 dilution respectively.. Blots were incubated with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (**ab216772**) and Goat anti-Rabbit IgG H&L (IRDye® 680RD)

preabsorbed (**ab216777**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human NF2 (Merlin) knockout HeLa cell lysate (ab257179)

Lane 1: Wild-type HeLa cell lysate 20 ug
Lane 2: NF2 knockout HeLa cell lysate 20 ug
Lanes 1 - 2: Merged signal (red and green). Green - **ab109244** observed at 60 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.
ab109244 was shown to react with NF2 in wild-type HeLa cells in western blot with loss of signal observed in NF2 knockout cell line **ab261796** (NF2 knockout cell lysate ab257179). Wild-type and NF2 knockout HeLa cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab109244** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4˚C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively.. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Sanger Sequencing - Human NF2 knockout HeLa cell lysate (ab257179)

Homozygous: 1 bp deletion in exon 1

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