abcam

ab52627

Anti-Notch1 antibody [EP1238Y]

Anti-Notch1 antibody [EP1238Y] (ab52627) is a rabbit monoclonal antibody detecting Notch1 in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF**. Suitable for **Human, Mouse**.

- KO validated for confirmed specificity
- Biophysical QC for unrivalled batch-batch consistency
- Over 200 publications
- Trusted since 2007

Recombinant | RabMAb | Advanced Validation | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EP1238Y
Purification technique	Affinity purification Protein A
Specificity	80% identities with Notch 2 and 81% with Notch 3
Concentration	1.123 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/100 - 1/150
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Not recommended

Species	Mouse, Cow
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100 - 1/150
Notes	-

Not recommended

Species	Mouse, Cow
Dilution info	-
Notes	-

IP

Not recommended

Species	Mouse, Human, Cow
Dilution info	-
Notes	-

WB

Tested

Species	Mouse
Dilution info	1/1000 - 1/2000
Notes	-

Species	Human
Dilution info	1/1000 - 1/2000
Notes	-

Predicted

Species	Cow
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/130 - 1/200
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Cow
Dilution info	-
Notes	-

ChIC/CUT&RUN-seq

Tested

Species	Human
Dilution info	0.5 µg
Notes	-

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Cow
Dilution info	-
Notes	-

Target data

See full target information NOTCH1

Function

Functions as a receptor for membrane-bound ligands Jagged-1 (JAG1), Jagged-2 (JAG2) and Delta-1 (DLL1) to regulate cell-fate determination. Upon ligand activation through the released notch intracellular domain (NICD) it forms a transcriptional activator complex with RBPJ/RBPSUH and activates genes of the enhancer of split locus. Affects the implementation of differentiation, proliferation and apoptotic programs. Involved in angiogenesis; negatively regulates endothelial cell proliferation and migration and angiogenic sprouting. Involved in the maturation of both CD4(+) and CD8(+) cells in the thymus. Important for follicular differentiation and possibly cell fate selection within the follicle. During cerebellar development, functions as a receptor for neuronal DNER and is involved in the differentiation of Bergmann glia. Represses neuronal and myogenic differentiation. May play an essential role in postimplantation development, probably in some aspect of cell specification and/or differentiation. May be involved in mesoderm development, somite formation and neurogenesis. May enhance HIF1A function by sequestering HIF1AN away from HIF1A. Required for the THBS4 function in regulating protective astrogenesis from the subventricular zone (SVZ) niche after injury. Involved in determination of left/right symmetry by modulating the balance between motile and immotile (sensory) cilia at the left-right organiser (LRO).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

Notch 1 (encoded by the Notch 1 gene) is a member of the Notch family of receptors. When bound to a ligand, the Notch1 receptor sends signals critical for normal development of many tissues throughout the body. Notch1 signalling supports the determination and specialization of cells as well as playing a role in cell proliferation, differentiation, and apoptosis.

Product Specifications
Anti-Notch1 antibody [EP1238Y] (ab52627) was developed by Abcam using patented rabbit monoclonal antibody technology and is validated for use in Flow Cyt (Intra), ICC/IF, IHC-P, WB in human, mouse samples.
Anti-Notch1 antibody [EP1238Y] (ab52627) specifically detects Notch1 (UniProt ID: P46531; Molecular weight: 271kDa) and is sold in 100 µL and 1 mL selling sizes.

Quality and Validation
Abcam's high quality manufacturing and validation processes ensure Anti-Notch1 antibody [EP1238Y] (ab52627) has high sensitivity and specificity alongside high lot-to-lot consistency and reproducibility.
The specificity of Anti-Notch1 antibody [EP1238Y] (ab52627) has been confirmed by testing in knockout samples.
Anti-Notch1 antibody [EP1238Y] (ab52627) has been cited over 201 times in peer reviewed journals and is trusted by the scientific community.
Anti-Notch1 antibody [EP1238Y] (ab52627) has 17 independent reviews from customers.

Related Products
Antibody clone EP1238Y is also available pre-conjugated to a variety of labels for your convenience - Alexa Fluor® 647, PE, APC, HRP, Alkaline Phosphatase, Alexa Fluor® 488, Alexa Fluor® 594, Alexa Fluor® 555, Alexa Fluor® 750 (ab194122, ab303041, ab303042, ab303043, ab308809, ab309758, ab310540, ab315420, ab321656).

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

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Notch1 also known as Notch-1 is a transmembrane receptor involved in cell fate decisions. It belongs to the Notch protein family and has a molecular weight of around 270 kDa. Notch1 is expressed widely particularly in tissues such as blood cells and neuronal tissue. The receptor consists of extracellular EGF-like repeats a transmembrane domain and an intracellular domain that translocates to the nucleus upon activation. Notch1 plays a critical role in intercellular communication through ligand engagement which then triggers a series of proteolytic cleavages that release the Notch intracellular domain (NICD) for nuclear translocation.
Biological function summary	Notch1 is essential in various cellular processes including differentiation proliferation and apoptosis. It functions as part of a complex signaling pathway that regulates these processes. Notch1 interacts with other elements like Jagged and Delta/Serrate/LAG-2 (DSL) family ligands to control gene expression patterns that determine cell lineage outcomes. This interaction affects the development of many systems such as the immune and nervous systems. Consequently Notch1 significantly influences the formation of organs and the maintenance of stem cell populations.
Pathways	Notch1 plays a significant role in both the Notch signaling and the Wnt signaling pathways. In the Notch signaling pathway Notch1 upon ligand binding partners with CSL (CBF1/RBP-Jκ in mammals) and Mastermind-like proteins for transcriptional regulation. This pathway interlinks with the Wnt pathway that involves proteins like β-catenin affecting the regulation of gene transcription. The interplay between Notch1 and these pathways is fundamental in determining outcomes in cell proliferation and differentiation emphasizing the interconnected nature of signaling networks.
Associated diseases and disorders	Notch1 is associated with T-cell acute lymphoblastic leukemia and breast cancer. Altered Notch1 signaling often through gain-of-function mutations can drive oncogenesis by disrupting normal cell differentiation and promoting uncontrolled proliferation. In T-cell acute lymphoblastic leukemia aberrant activation of Notch1 leads to increased expression of target genes working closely with related proteins like c-Myc. In breast cancer dysregulation of Notch1 signaling may facilitate tumor growth and metastasis indicating its role as a therapeutic target.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

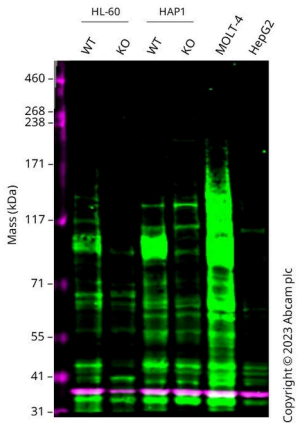
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

---

19 product images



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Western blot: Anti-NOTCH1 antibody [EP1238Y] (ab52627) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab52627 was shown to bind specifically to NOTCH1. A band was observed at 110-120 kDa in wild-type HL-60 cell lysates with no signal observed at this size in NOTCH1 knockout cell line. To generate this image, wild-type and NOTCH1 knockout HL-60 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 IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.
All lanes:
Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627) at 1/1000 dilution

Lane 1:
Wild-type HL-60 cell lysate at 20 µg

Lane 2:
NOTCH1 knockout HL-60 cell lysate at 20 µg

Lane 3:
Wild-type HAP1 cell lysate at 20 µg

Lane 4:
NOTCH1 knockout HAP1 cell lysate at 20 µg

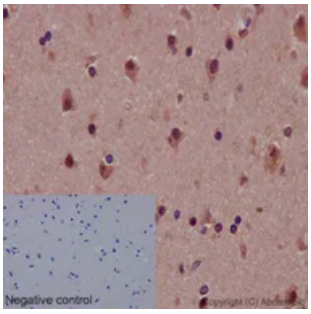
Lane 5:
MOLT-4 cell lysate at 20 µg

Lane 6:
HepG2 cell lysate at 20 µg

Developed using the ECL technique.

Performed under reducing conditions.

Observed band size: 110-120 kDa

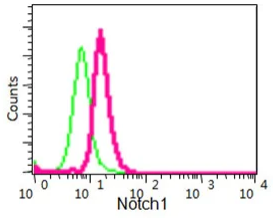


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Notch1 antibody

Immunohistochemical staining of paraffin-embedded human brain with purified ab52627 at a dilution of 1/150.
A prediluted HRP polymer for rabbit IgG was used as the secondary and the sample was stained with hematoxylin. PBS was used instead of the primary antibody as the **negative control**, and is shown in the inset.

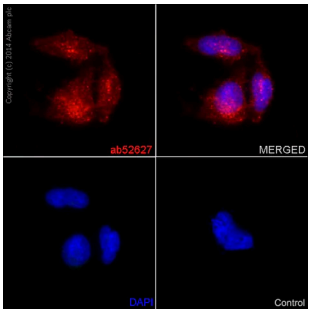
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Flow Cytometry (Intracellular) staining using rabbit Anti-Notch1 antibody

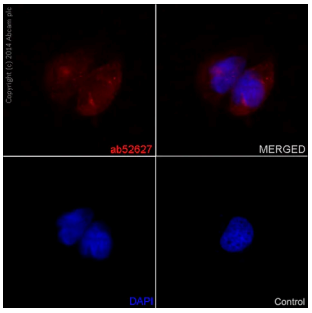
Intracellular Flow Cytometry analysis of permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells (2% PFA, pink) withpurified ab52627 at a 1/200 dilution, or **negative control** rabbit monoclonal IgG (green). The secondary antibody was FITC goat anti-rabbit.



Immunocytochemistry/ Immunofluorescence - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Notch1 antibody

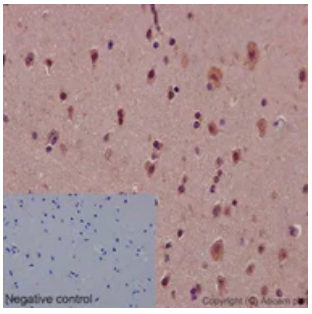
Immunofluorescent staining of HeLa (Human epithelial cell line from cervix adenocarcinoma) cells fixed with 4% PFA using purified ab52627 at a dilution of 1/150.
An Alexa Fluor® 555 goat anti-rabbit was used as the secondary and the sample was stained with DAPI. An Alexa Fluor® 555 goat anti-rabbit was used at a dilution of 1/500 as the **negative control** and is shown in the bottom right hand panel.



Immunocytochemistry/ Immunofluorescence - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Notch1 antibody

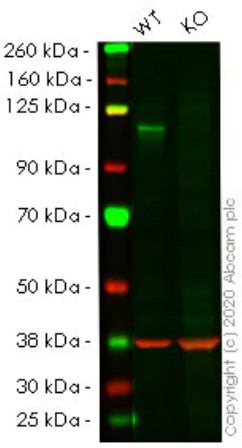
Immunofluorescent staining of HeLa (Human epithelial cell line from cervix adenocarcinoma) cells fixed with 4% PFA using unpurified ab52627 at a dilution of 1/100.
An Alexa Fluor® 555 goat anti-rabbit was used as the secondary and the sample was stained with DAPI. An Alexa Fluor® 555 goat anti-rabbit was used at a dilution of 1/500 as the **negative control** and is shown in the bottom right hand panel.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Notch1 antibody [EP1238Y] (ab52627)

Immunohistochemical staining of paraffin-embedded human brain with unpurified ab52627 at a dilution of 1/100.
A prediluted HRP polymer for rabbit IgG was used as the secondary and the sample was stained with hematoxylin. PBS was used instead of the primary antibody as the **negative control**, and is shown in the inset.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining using rabbit Anti-Notch1 antibody

Lanes 1- 2: Merged signal (red and green). Green - ab52627 observed at 110-120 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.
ab52627 was shown to react with Notch1 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab261762 (knockout cell lysate ab257006) was used. Wild-type HeLa and NOTCH1 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab52627 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627) at 1/1000 dilution

Lane 1:
Wild-type HeLa cell lysate at 20 µg

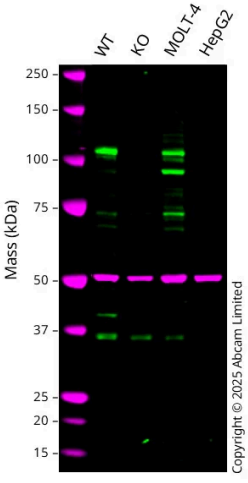
Lane 2:
Western blot - Human NOTCH1 knockout HeLa cell line (ab261762)

Lane 2:
NOTCH1 knockout HeLa cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 272 kDa

Observed band size: 110-120 kDa



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining using rabbit Anti-Notch1 antibody

Western blot: Rabbit Monoclonal [EP1238Y] to Notch1 ab52627 staining at 1/1000 dilution, shown in green; Mouse anti alpha Tubulin ab7291 loading control staining at 1/20000 dilution, shown in magenta. A band was observed at 105 kDa in Wild-type HCT 116 cell lysates with no signal observed at this size in NOTCH1 knockout HCT 116 cell line. To generate this image, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 5pc 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 800CW and Goat anti-Mouse 680RD at 1/20,000 dilution.
All lanes:
Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627) at 1/1000 dilution

Lane 1:
Wild-type HCT 116 whole cell lysate at 20 µg

Lane 2:
Western blot - Human NOTCH1 knockout HCT116 cell line (ab287652) at 20 µg

Lane 3:
MOLT-4 whole cell lysate at 20 µg

Lane 4:
HepG2 whole cell lysate at 20 µg

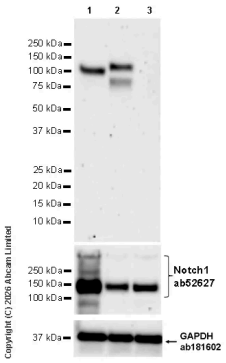
Secondary

All lanes:
Goat anti-Rabbit 800CW & Goat anti-Mouse 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 100 kDa

Observed band size: 105 kDa



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining using rabbit Anti-Notch1 antibody

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

Negative control: MCF7

To minimize protein degradation, cells were lysed immediately after harvest and lysates applied to a gel and transfer membrane for Western blotting as soon as possible.The identity of the lower MW bands are likely to be degradation fragments.

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) (1:200000) (36KDa).

All lanes:
Western blot - Anti-Cleaved Notch1 (Val1744) antibody [EPR26110-518] (ab327383) at 1/1000 dilution

Lane 1:
Jurkat (human T cell leukemia T lymphocyte from peripheral blood) whole cell lysate at 40 µg

Lane 2:
MOLT-4 (human lymphoblastic leukemia T lymphoblast) whole cell lysate at 40 µg

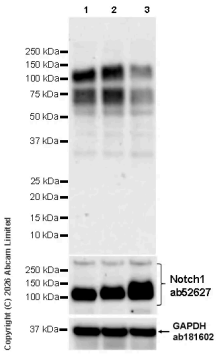
Lane 3:
MCF7 (human breast adenocarcinoma epithelial cell) whole cell lysate at 40 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Observed band size: 110 kDa, 36 kDa, 300 kDa

Exposure time: 180s



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining using rabbit Anti-Notch1 antibody

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

DAPT is a γ-secretase inhibitor that blocks the cleavage and subsequent release of the Notch1 intracellular domain (NICD).

To minimize protein degradation, cells were lysed immediately after harvest and lysates applied to a gel and transfer membrane for Western blotting as soon as possible.The identity of the lower MW bands are likely to be degradation fragments.

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) (1:200000) (36KDa).

All lanes:
Western blot - Anti-Cleaved Notch1 (Val1744) antibody [EPR26110-518] (ab327383) at 1/1000 dilution

Lane 1:
Untreated Jurkat (human T cell leukemia T lymphocyte from peripheral blood) whole cell lysate at 40 µg

Lane 2:
Jurkat treated with 0.1 µM DAPT for 24 hours whole cell lysate at 40 µg

Lane 3:
Jurkat treated with 1 µM DAPT for 24 hours whole cell lysate at 40 µg

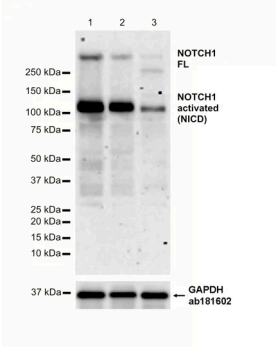
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Developed using the ECL technique.

Observed band size: 110 kDa, 36 kDa, 300 kDa

Exposure time: 180s



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining using rabbit Anti-Notch1 antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST.
In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

All lanes:
Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627) at 1/1000 dilution

Lane 1:
HeLa (human cervical adenocarcinoma epithelial cell)

Lane 2:
293T (human embryonic kidney epithelial cell)

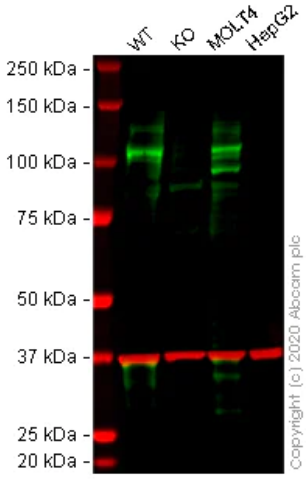
Lane 3:
NIH/3T3 (mouse embryonic fibroblast)

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 110 kDa, 272 kDa, 36 kDa

Exposure time: 180s



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining using rabbit Anti-Notch1 antibody

Lanes 1 - 4: Merged signal (red and green). Green - ab52627 observed at 110-120 kDa. Red - loading control, ab8245 (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.
ab52627 was shown to react with Notch1 in wild-type HAP1 cells in western blot. Loss of signal was observed when NOTCH1 knockout sample was used. Wild-type and NOTCH1 knockout HAP1 cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk before incubation with ab52627 and ab8245 (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at 1 µg/ml 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.

All lanes:
Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627) at 1 µg/mL

Lane 1:
Wild-type HAP1 cell lysate at 40 µg

Lane 2:
NOTCH1 knockout HAP1 cell lysate at 40 µg

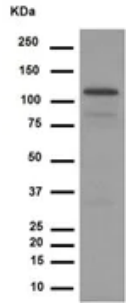
Lane 3:
MOLT-4 cell lysate at 20 µg

Lane 4:
HepG2 cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 272 kDa

Observed band size: 110-120 kDa



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627) at 1/2000 dilution

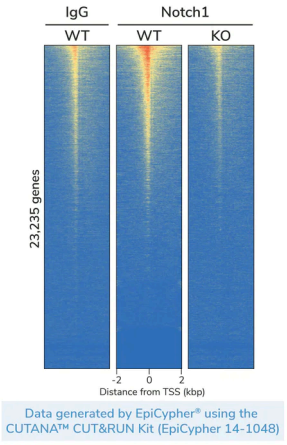
All lanes:
HEK-293 (Human epithelial cell line from embryonic kidney) cell lysate at 10 µg

Secondary

All lanes:
HRP goat anti-rabbit (H+L) at 1/1000 dilution

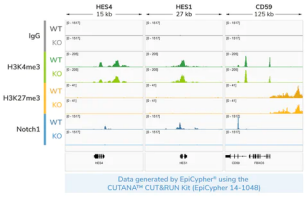
Predicted band size: 272 kDa

Observed band size: 110-120 kDa



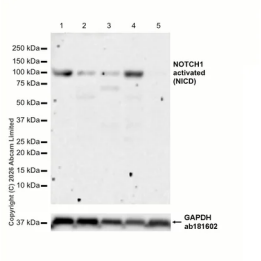
ChIC/CUT&RUN sequencing - Anti-Notch1 antibody [EP1238Y] (ab52627)

CUT&RUN profiling with Notch1 antibody demonstrates robust genome-wide enrichment in wild-type (WT) cells, which is markedly diminished in NOTCH1 knockout (KO) cells. Heatmaps of genome-wide signal flanking annotated transcription start sites (TSSs, +/- 2 kbp) display CUT&RUN data generated using the CUTANA™ CUT&RUN Kit (EpiCypher 14-1048) with Notch1 antibody (Abcam ab52627, 0.5 µg). 500,000 HeLa WT or KO (Abcam ab261762) cells were used per reaction. IgG antibody was included as a negative control to assess non-specific background. Libraries were prepared using the CUTANA™ CUT&RUN Library Prep Kit (EpiCypher 14-1001). Sequencing was performed with paired-end 50 bp reads, and data were processed on CUTANA™ Cloud (cloud.epicypher.com) by alignment to the hg38 genome. Heatmaps were generated using deepTools (Ramнrez et al., Nucleic Acids Res. 2014; PMID: 24799436). Row-linked data are ranked by intensity relative to Notch1 WT, with red indicating high localized enrichment and blue denoting background. Validated antibodies show genome-wide enrichment above IgG background consistent with Notch1 binding in WT cells and near complete loss of signal in KO cells.



ChIC/CUT&RUN sequencing - Anti-Notch1 antibody [EP1238Y] (ab52627)

CUT&RUN profiling with Notch1 antibody reveals the expected genomic enrichment pattern in wild-type (WT) cells, which is substantially reduced in NOTCH1 knockout (KO) cells. Representative genome browser tracks show CUT&RUN data generated using the CUTANA™ CUT&RUN Kit (EpiCypher 14-1048) with Notch1 antibody (Abcam ab52627, 0.5 µg). 500,000 HeLa WT or KO (ab261762) cells were used per reaction. IgG, H3K4me3, and H3K27me3 antibodies were included as controls to assess non-specific background, active promoters, and repressed chromatin, respectively. Libraries were prepared using the CUTANA™ CUT&RUN Library Prep Kit (EpiCypher 14-1001). Sequencing was performed with paired-end 50 bp reads, and data were processed on CUTANA™ Cloud (cloud.epicypher.com) by alignment to the hg38 genome. Images were generated using Integrative Genomics Viewer (IGV, Broad Institute).



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining using rabbit Anti-Notch1 antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST.
In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

Low expression: Ovary.

All lanes:
Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627) at 1/1000 dilution

Lane 1:
Mouse brain tissue lysate at 40 µg

Lane 2:
Mouse heart tissue lysate at 40 µg

Lane 3:

Mouse spleen tissue lysate at 40 µg

Lane 4:
Mouse lung tissue lysate at 40 µg

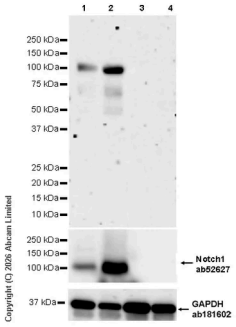
Lane 5:
Mouse ovary tissue lysate at 40 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 110 kDa, 36 kDa

Exposure time: 180s



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining using rabbit Anti-Notch1 antibody

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

Low expression: EL4, Neuro-2a.

To minimize protein degradation, cells were lysed immediately after harvest and lysates applied to a gel and transfer membrane for Western blotting as soon as possible.The identity of the lower MW bands are likely to be degradation fragments.

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) (1:200000) (36KDa).

All lanes:
Western blot - Anti-Cleaved Notch1 (Val1744) antibody [EPR26110-518] (ab327383) at 1/1000 dilution

Lane 1:
HC11 (mouse mammary gland epithelial cell) whole cell lysate at 40 µg

Lane 2:
bEnd.3 (mouse brain endothelial cell) whole cell lysate at 40 µg

Lane 3:
EL4 (mouse lymphoma T lymphocyte) whole cell lysate at 40 µg

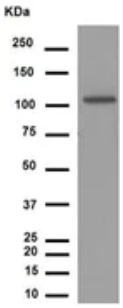
Lane 4:
Neuro-2a (mouse neuroblastoma neuroblast) whole cell lysate at 40 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Observed band size: 110 kDa, 36 kDa, 300 kDa

Exposure time: 180s



Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627)

Notch1 Western blot staining of Mouse brain using rabbit Anti-Notch1 antibody

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-Notch1 antibody [EP1238Y] (ab52627) at 1/2000 dilution

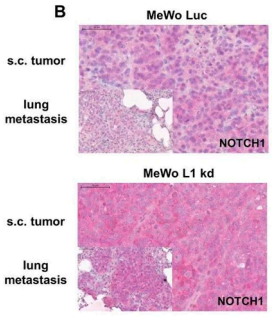
All lanes:
Mouse brain at 10 µg

Secondary

All lanes:
HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 272 kDa

Observed band size: 110-120 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Notch1 antibody [EP1238Y] (ab52627)

Verification of gene expression array data by immunohistochemical analysis of Notch 1 expression in subcutaneous tumors and lung metastases from a human melanoma (MeWo) xenograft experiment in mice.
Immunohistochemical staining for Notch1 expression (ab52627, red) in subcutaneous tumors and lung metastases (both panels) of MeWo (Human malignant melanoma cell line) cells.

All scale bars: 50 µm.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.