


## Product datasheet

### Anti-NOTCH3 antibody ab23426

★★★★★ [22 Abreviews](#) [118 References](#) [6 Images](#)

#### Overview

<b>Product name</b>	Anti-NOTCH3 antibody
<b>Description</b>	Rabbit polyclonal to NOTCH3
<b>Host species</b>	Rabbit
<b>Specificity</b>	From Jan 2024, QC testing of replenishment batches of this polyclonal changed. All tested and expected application and reactive species combinations are still covered by our Abcam product promise. However, we no longer test all applications. For more information on a specific batch, please contact our Scientific Support who will be happy to help.
<b>Tested applications</b>	<b>Suitable for:</b> WB, ICC/IF, IHC-P
<b>Species reactivity</b>	<b>Reacts with:</b> Mouse, Human <b>Predicted to work with:</b> Rat 
<b>Immunogen</b>	Synthetic peptide corresponding to Human NOTCH3 aa 2300 to the C-terminus (C terminal) conjugated to keyhole limpet haemocyanin. (Peptide available as <a href="#">ab26878</a> )
<b>Positive control</b>	ICC: mES cells, MCF7 cells, COS1 fibroblast cell line. WB: HeLa, K-562, Caco-2 cell lines. IHC-P: Human breast cancer and human breast carcinoma tissues.
<b>General notes</b>	<p>The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing.</p> <p>If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&amp;As</p>

#### Properties

<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
<b>Storage buffer</b>	pH: 7.40 Preservative: 0.02% Sodium azide Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising agent. If you would like information about the formulation of a specific lot, please contact our scientific support team who will be happy to help.

<b>Purity</b>	Immunogen affinity purified
<b>Clonality</b>	Polyclonal
<b>Isotype</b>	IgG

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab23426 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	★★★★★ (6)	Use a concentration of 1 µg/ml. Detects a band of approximately 97, 280 kDa (predicted molecular weight: 244 kDa).
ICC/IF	★★★★★ (3)	Use a concentration of 1 - 5 µg/ml.
IHC-P	★★★★★ (9)	Use at an assay dependent concentration.

## Target

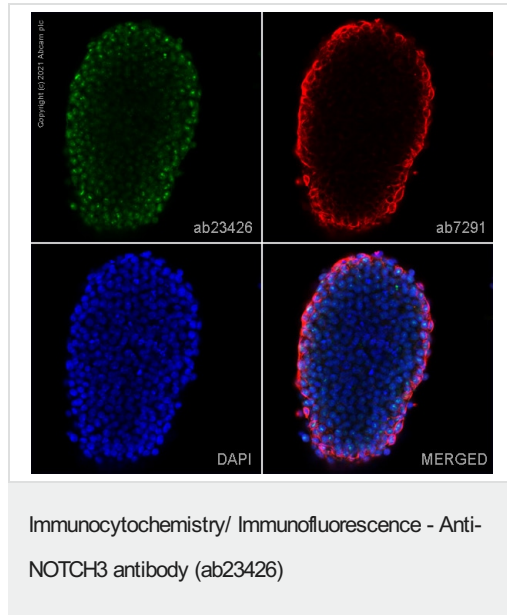
<b>Function</b>	Functions as a receptor for membrane-bound ligands Jagged1, Jagged2 and Delta1 to regulate cell-fate determination. Upon ligand activation through the released notch intracellular domain (NICD) it forms a transcriptional activator complex with RBPJ/RBPSUH and activates genes of the enhancer of split locus. Affects the implementation of differentiation, proliferation and apoptotic programs.
<b>Tissue specificity</b>	Ubiquitously expressed in fetal and adult tissues.
<b>Involvement in disease</b>	Defects in NOTCH3 are the cause of cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL) [MIM:125310]. CADASIL causes a type of stroke and dementia of which key features include recurrent subcortical ischemic events and vascular dementia. The disorder affects relatively young adults of both sexes. Mutations affect highly conserved cysteine residues within epidermal growth factor (EGF)-like repeat domains in the extracellular part of the receptor.
<b>Sequence similarities</b>	Belongs to the NOTCH family. Contains 5 ANK repeats. Contains 34 EGF-like domains. Contains 3 LNR (Lin/Notch) repeats.
<b>Post-translational modifications</b>	Synthesized in the endoplasmic reticulum as an inactive form which is proteolytically cleaved by a furin-like convertase in the trans-Golgi network before it reaches the plasma membrane to yield an active, ligand-accessible form. Cleavage results in a C-terminal fragment N(TM) and a N-terminal fragment N(EC). Following ligand binding, it is cleaved by TNF-alpha converting enzyme (TACE) to yield a membrane-associated intermediate fragment called notch extracellular truncation (NEXT). This fragment is then cleaved by presenilin dependent gamma-secretase to release a notch-derived peptide containing the intracellular domain (NICD) from the membrane.

Phosphorylated.

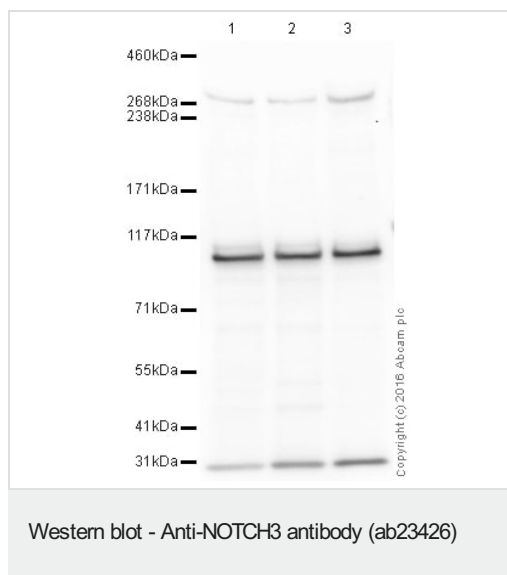
## Cellular localization

Cell membrane and Nucleus. Following proteolytical processing NICD is translocated to the nucleus.

## Images



ab23426 staining NOTCH3 in mES cells. The cells were fixed with 4% paraformaldehyde (10 min), permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab23426 at 1µg/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with **ab150081**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150120**, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue). Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.



**All lanes** : Anti-NOTCH3 antibody (ab23426) at 1 µg/ml

**Lane 1** : K-562 whole cell lysate (**ab29306**)

**Lane 2** : Caco-2 whole cell lysate (**ab3950**)

**Lane 3** : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

### Secondary

**All lanes** : Goat polyclonal to Rabbit IgG - H&L (HRP) at 1/50000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size:** 244 kDa

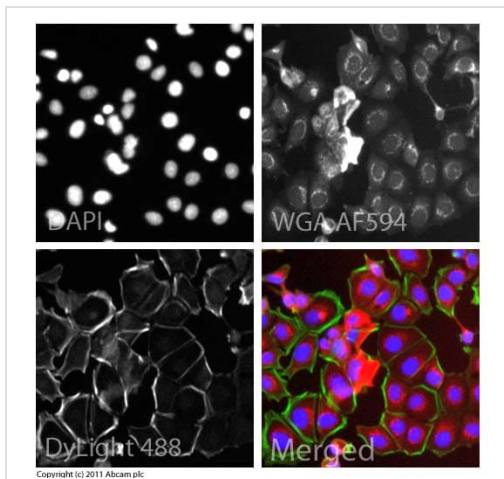
**Observed band size:** 280,97 kDa

**Additional bands at:** 105 kDa, 30 kDa. We are unsure as to the identity of these extra bands.

**Exposure time:** 30 seconds

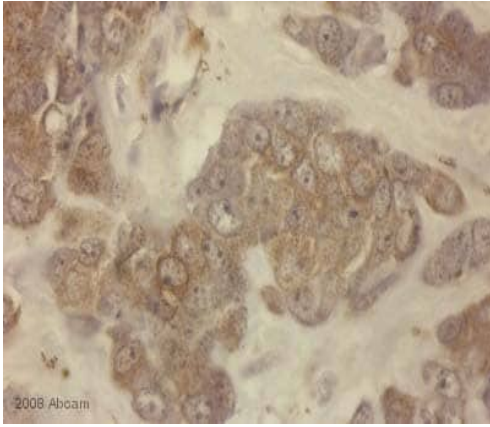
This blot was produced using a 3-8% Tris Acetate gel under the TA buffer system. The gel was run at 150V for 60 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 5% Bovine Serum Albumin before being incubated with ab23426 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution.

The band observed at 97 kDa is thought to correspond to the notch-derived peptide containing the intracellular domain (NICD) of NOTCH3 as described in the literature (PMID:10712431).



ICC/IF image of ab23426 stained MCF7 cells. The cells were 100% methanol fixed (5 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 (ab23426, 5µg/ml) overnight at +4°C. The secondary antibody (green) was **ab96899**, DyLight® 488 goat anti-rabbit 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.

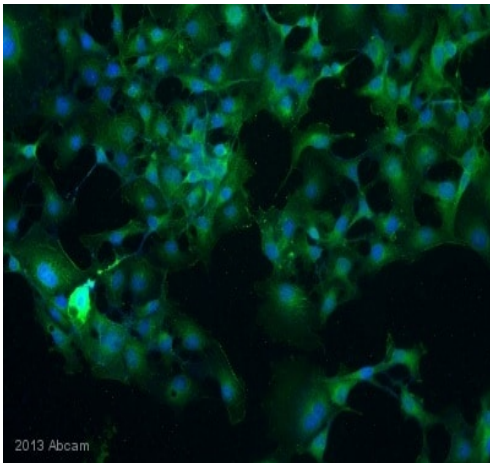
Immunocytochemistry/ Immunofluorescence - Anti-  
NOTCH3 antibody (ab23426)



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NOTCH3 antibody (ab23426)

This image is courtesy of an Abreview submitted by Antibody Solutions Ltd.

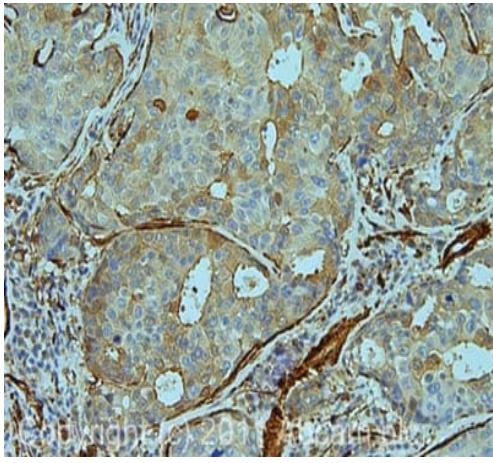
ab23426 staining NOTCH3 in human breast cancer tissue sections by immunohistochemistry (Formalin/PFA-fixed paraffin embedded sections). Tissue underwent fixation in paraformaldehyde, heat-mediated antigen retrieval in citrate buffer pH6.0 and blocking for 15 minutes at 20°C (5 minutes for peroxidase blocking and 10 minutes for protein blocks). The primary antibody was diluted 1/250 and incubated with sample for 45 minutes at 20°C. A HRP-conjugated goat polyclonal to rabbit IgG was used undiluted as secondary.



Immunocytochemistry/ Immunofluorescence - Anti-NOTCH3 antibody (ab23426)

This image is courtesy of an anonymous Abreview

ab23426 staining NOTCH3 in the COS1 fibroblast cell line from Monkey Kidney by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with paraformaldehyde, permeabilized with Triton X-100 0.1% in PBS and blocked with 1% BSA for 30 minutes at 25°C. Samples were incubated with primary antibody (1/200) for 16 hour at 4°C. An Alexa Fluor® 488-conjugated Goat anti-rabbit polyclonal (1/500) was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NOTCH3 antibody (ab23426)

IHC image of NOTCH3 staining in human breast carcinoma 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 ab23426, 5µg/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.

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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