

Product datasheet

Anti-NOTCH3 antibody ab23426

★★★★★ 14 Abreviews 39 References 7 图像

概述

产品名称	Anti-NOTCH3抗体
描述	兔多克隆抗体to NOTCH3
经测试应用	适用于: WB, IHC-P, ICC/IF, IHC-FoFr, IHC-Fr
种属反应性	与反应: Mouse, Rat, Human
免疫原	Synthetic peptide corresponding to Human NOTCH3 aa 2300 to the C-terminus (C terminal) conjugated to Keyhole Limpet Haemocyanin (KLH). (Peptide available as ab26878)
阳性对照	This antibody gave a positive signal in the following Human Cell lysates: K562 and CACO-2

性能

形式	Liquid
存放说明	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.
存储溶液	Preservative: 0.02% Sodium Azide Constituents: 1% BSA, PBS, pH 7.4
纯度	Immunogen affinity purified
克隆	多克隆
同种型	IgG

应用

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.

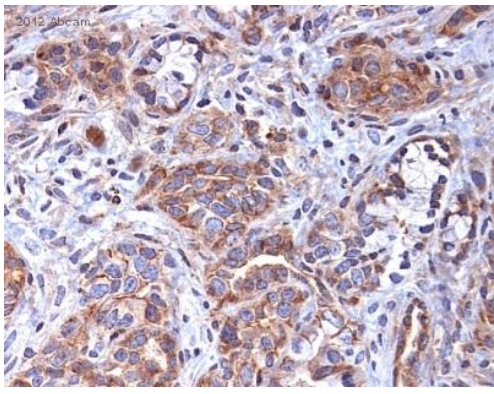
应用	Ab评论	说明
WB	★★★★★	Use a concentration of 1 µg/ml. Detects a band of approximately 97,280 kDa (predicted molecular weight: 244 kDa).

应用	Ab评论	说明
IHC-P	★★★★★	Use a concentration of 5 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. PubMed: 19965627
ICC/IF	★★★★★	Use at an assay dependent concentration. PubMed: 20680961
IHC-FoFr	★★★★☆	Use at an assay dependent concentration.
IHC-Fr	★★★★★	1/300.

靶标

功能	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.
组织特异性	Ubiquitously expressed in fetal and adult tissues.
疾病相关	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.
序列相似性	Belongs to the NOTCH family. Contains 5 ANK repeats. Contains 34 EGF-like domains. Contains 3 LNR (Lin/Notch) repeats.
翻译后修饰	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.
细胞定位	Cell membrane and Nucleus. Following proteolytical processing NICD is translocated to the nucleus.

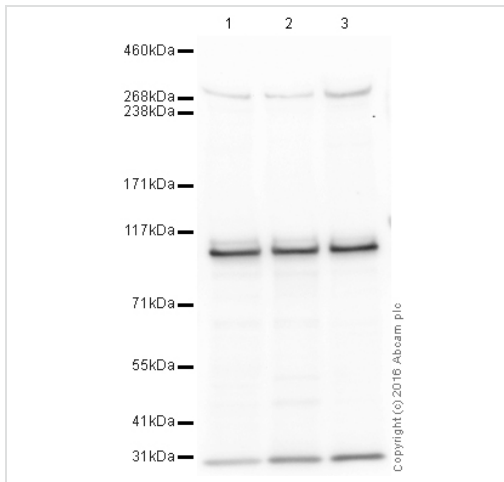
图片



Immunohistochemistry (Frozen sections) - Anti-
NOTCH3 antibody (ab23426)

This image is courtesy of an anonymous Abreview

ab23426 staining NOTCH3 in mouse pancreatic cancer tissue sections by Immunohistochemistry (IHC-Fr - frozen sections). Tissue was fixed with acetone and blocked with 10% serum for 1 hour at 20°C. Samples were incubated with the primary antibody (1/100 in PBS) for 8 hours at 4°C. A biotin-conjugated goat anti-rabbit IgG polyclonal (1/1000) was used as the secondary antibody.



Western blot - Anti-NOTCH3 antibody (ab23426)

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

Lane 1 : K562 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

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

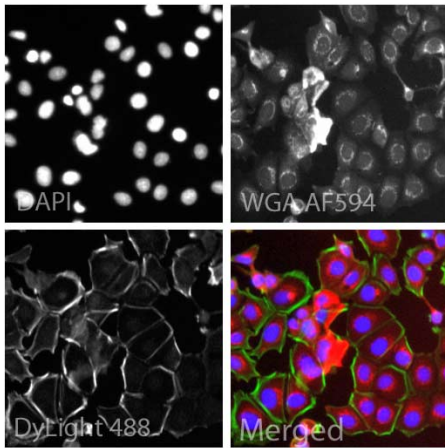
Observed band size : 97,280 kDa

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

Exposure time : 30 seconds

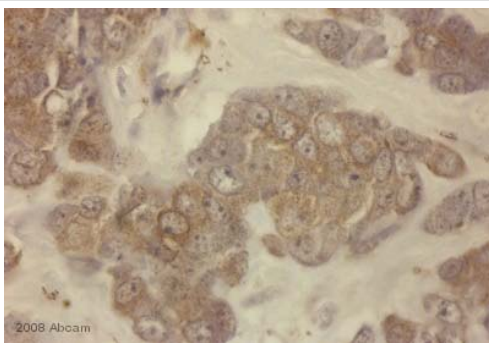
This blot was produced using a 3-8% Tris Acetate gel under the TA buffer system. The gel was run at 150V for 60 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 5% Bovine Serum Albumin before being incubated with ab23426 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).



Immunocytochemistry/ Immunofluorescence - Anti-NOTCH3 antibody (ab23426)

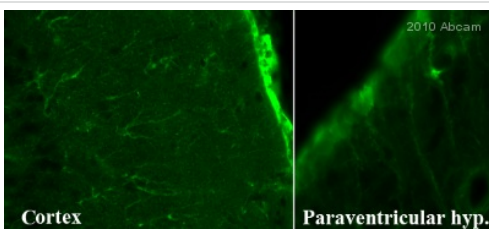
ICC/IF image of ab23426 stained MCF7 cells. The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (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.



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

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

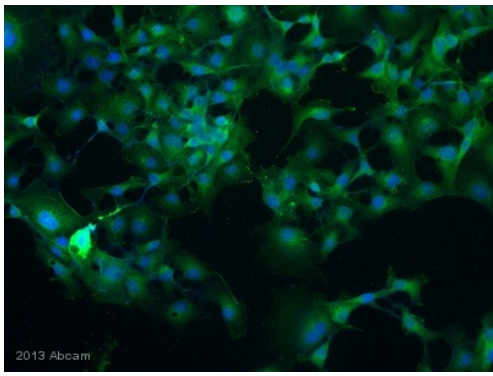
ab23426 staining NOTCH3 in human breast cancer tissue sections by immunohistochemistry (Formalin/PFA-fixed paraffin embedded sections). Tissue underwent fixation in paraformaldehyde, heat-mediated antigen retrieval in citrate buffer pH6.0 and blocking for 15 minutes at 20°C (5 minutes for peroxidase 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.



Immunohistochemistry (PFA perfusion fixed frozen sections) - Anti-NOTCH3 antibody (ab23426)

Image courtesy of Sophie Pezet, ESPCI, France

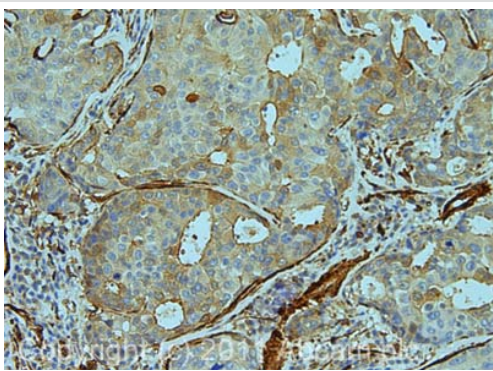
IHC-FoFr image of NOTCH3 staining in Rat cerebral cortex (left) and the paraventricular hypothalamic nucleus (right) sections. The sections used came from animals perfused fixed with Paraformaldehyde 4%, in phosphate buffer 0.2M. Following postfixation in the same fixative overnight, the brains were cryoprotected in sucrose 30% overnight. Immunostainings were performed using the 'free floating' technique.



Immunocytochemistry/ Immunofluorescence - Anti-NOTCH3 antibody (ab23426)

This image is courtesy of an anonymous Abreview

ab14404 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 hours 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.

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