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Datasheet for 100-401-408

NOTCH 2 Antibody

Overview

Description:	Anti-NOTCH 2 (Cleaved N terminal) (Human specific) (RABBIT) Antibody - 100-401-408
Item No.:	100-401-408
Size:	200 µL
Applications:	ELISA, IHC, WB
Reactivity:	Human
Host Species:	Rabbit

Product Details

Background:	Anti-Notch 2 Antibody recognizes Notch2 that is synthesized in the endoplasmic reticulum as an inactive form which is proteolytically cleaved by a furin-like convertase (S1 cleavage) 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 (S2 cleavage) 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 (S3 cleavage) to release the intracellular domain (NICD) from the membrane. Anti-NOTCH2 Antibody is useful for researchers interested in Notch1, Jagged1, Jagged2, and Delta1, as well as neuroscience, transcription and cancer research.
Synonyms:	rabbit anti-Notch2 antibody, AGS2 antibody, hN2 antibody, Notch homolog 2 antibody, neurogenic locus notch homolog protein 2, NICD
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	Antiserum

Target Details

Gene Name:	NOTCH2
Reactivity:	Human
Immunogen Type:	Conjugated Peptide

Immunogen:	This whole rabbit serum was prepared by repeated immunizations with a synthetic peptide corresponding to amino acid residues of human Notch 2 located near the N-terminal sequence of the cleaved N intracellular domain (NICD).
Purity/Specificity:	This antiserum is directed against human NOTCH2. The immunogen epitope is only exposed after gamma secretase cleavage and is not accessible in the uncleaved form. Based on sequences, we expect this antibody to react as well with rat and mouse NOTCH 2. Only one amino acid difference (conservative) occurs in mouse and rat.
Relevant Links:	• UniProtKB - Q04721 • NCBI - 24041035 • GeneID - 4853

Application Details

Tested Applications:	ELISA, IHC, WB
Application Note:	Anti-Notch2 has been tested in ELISA, WB, and IHC. Anti-NOTCH-2 has a strong response was detected by ELISA against the immunizing peptide. This product was assayed against the peptide immunogen in a standard capture ELISA using Peroxidase conjugated anti-Rabbit IgG [H&L] (Goat) (code #611-1302) and ABTS (2,2'-azino-bis-[3-ethylbenthiazoline-6-sulfonic acid]) (code # ABTS-100) as a substrate for 30 minutes at room temperature. A working dilution of 1:30,000 to 1:90,000 is suggested in ELISA this product.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:20,000 - 1:100,000
IHC:	1:500
WB:	1:400 - 1:2,000

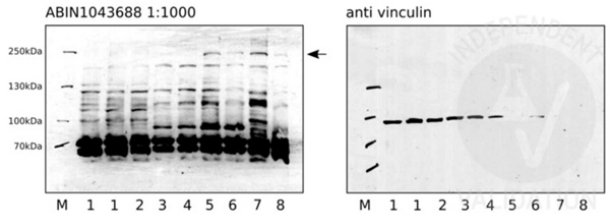
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	75 mg/mL by Refractometry
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	None

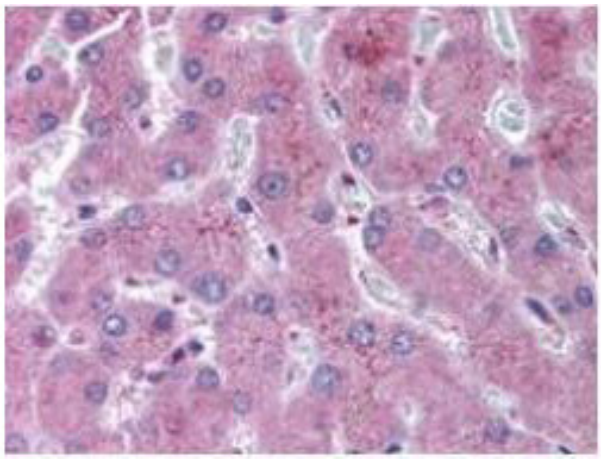
Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
Expiration:	Expiration date is one (1) year from date of receipt.

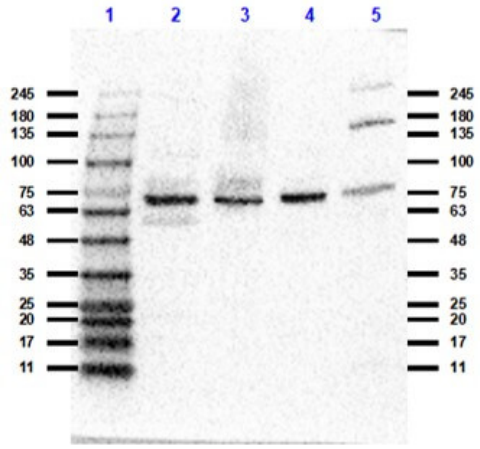
Images



Western Blot
Western blot Rabbit Anti-NOTCH 2 (Cleaved N terminal) Antibody.
Negative Control: human pluripotent stem cells (lanes 1 and 2), definite endoderm cells (lanes 3 and 4);
Positive Control: pancreatic endoderm cells (lanes 5 and 6), pancreatic progenitors cells (lanes 7 and 8).
Primary Antibody: Anti-NOTCH 2 used at a 1:1000 dilution overnight at 4°C. Vinculin served as loading control at 1:1000 at RT for 1hr.
Secondary Antibody: donkey anti-rabbit HRP conjugated antibody or donkey anti-mouse HRP conjugated antibody diluted 1:5000 for 1h at RT.
Strong unspecific bands. Nevertheless, a band over 250kDa appears to be highly specific, as it is only detected at day 10 (lanes 5 and 6) and day 14 (lanes 7 and 8).
Independently Validated by antibodies-online GmbH (p/n ABIN1043688/ ABIN104899) courtesy of Ulm University Hospital.

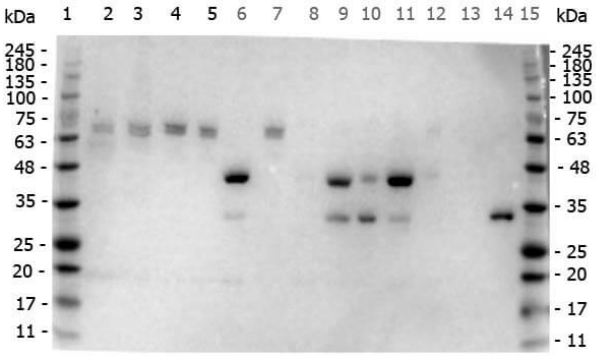


Immunohistochemistry
Rockland's anti-NOTCH 2 antibody was diluted 1:500 to detect NOTCH 2 in human liver tissue. Tissue was formalin fixed and paraffin embedded. No pre-treatment of sample was required. The image shows the localization of antibody as the precipitated red signal, with a hematoxylin purple nuclear counter stain.



Western Blot

Western Blot of Rabbit Anti-Notch 2 N-Term Antibody. Lane 1: Opal Prestained Molecular Weight Marker (p/n MB-210-0500). Lane 2: HeLa Whole Cell Lysate (p/n W09-000-364). Lane 3: MOLT-4 Whole Cell Lysate (p/n W09-001-GK4). Lane 4: K-562 Whole Cell Lysate (p/n W09-001-GJ7). Lane 5: Human Kidney Lysate. Load: 20µg lysate/lane. Primary Antibody: Anti-NOTCH 2 at 1:1000 for 1 hr at RT. Secondary Antibody: Goat Anti-Rabbit IgG HRP (p/n 611-103-122) at 1:70,000 for 30mins at RT. Blocking: 5% BLOTTO. Expected MW: ~86kDa.



Western Blot

Western Blot of Rabbit anti-NOTCH2 (Cleaved N terminal) antibody. Lane 1 Marker: Opal Pre-stained ladder (p/n MB-210-0500). Lane 2: U-251 Lysate (p/n W09-001-GY4). Lane 3: PC-3 Lysate (p/n W09-001-GV6). Lane 4: MCF-7 Lysate (p/n W09-000-360). Lane 5: Hep-G2 Lysate (p/n W09-001-GJ5). Lane 6: A431 Lysate (p/n W09-000-361). Lane 7: A549 Lysate (p/n W09-001-372). Lane 8: J774A.1 Lysate (p/n W10-001-GX3). Lane 9: C2C12 Lysate (p/n W10-001-GL7). Lane 10: Mouse Embryonic Fibroblast Lysate (p/n W10-001-371). Lane 11: NIH/3T3 Lysate (p/n W10-000-358). Lane 12: PC-12 Lysate (p/n W12-001-GL9). Lane 13: Rat Brain Lysate (p/n W12-000-T077). Lane 14: Mouse Brain Lysate (p/n W10-000-T004). Lane 15 Marker: Opal Pre-stained Ladder. Load: 10 µg of lysate/lane. Primary antibody: NOTCH2 antibody at 1µg/mL overnight at 4°C. Secondary antibody: Peroxidase goat anti-rabbit secondary antibody (p/n 611-103-122) at 1:70,000 for 60 min at RT. Blocking Buffer: 1% Casein-TTBS for 30 min at RT. Predicted size: ~86kDa in human cell lines (2-7) for NOTCH2.

References

- Iervolino A et al. Potassium depletion induces cellular conversion in the outer medullary collecting duct altering Notch signaling pathway. *Sci Rep.* (2020)
- Guilmeau S et al. Intestinal deletion of Pofut1 in the mouse inactivates notch signaling and causes enterocolitis. *Gastroenterology.* (2008)

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