Product Datasheet

Niemann-Pick type C1 Like-1 Antibody - BSA Free NB400-127

Unit Size: 0.1 ml

Store at 4C. Do not freeze.

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NB400-127

Niemann-Pick type C1 Like-1 Antibody - BSA Free

Product Information	
Unit Size	0.1 ml
Concentration	1.0 mg/ml
Storage	Store at 4C. Do not freeze.
Clonality	Polyclonal
Preservative	0.1% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	Tris-Citrate/Phosphate (pH 7.0 - 8.0)
Target Molecular Weight	140 kDa
Product Description	
Host	Rabbit
Gene ID	29881
Gene Symbol	NPC1L1
Species	Human, Mouse, Rat, Porcine
Immunogen	A synthetic peptide made to an internal region of rat Niemann-Pick type C1 Like- 1 (between residues 1000-1100). [UniProt# Q6T3U3]
Product Application Details	
Applications	Western Blot, ELISA, Electron Microscopy, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Knockdown Validated
Recommended Dilutions	Western Blot 2 ug/ml, ELISA reported in scientific literature (PMID 21051527), Immunohistochemistry 1:100-1:250, Immunocytochemistry/ Immunofluorescence 2 ug/ml, Immunohistochemistry-Paraffin 1:100-1:250, Electron Microscopy reported in scientific literature (PMID 21051527), Knockdown Validated reported in scientific literature (PMID 35023619)
Application Notes	In Western blot a band is observed at ~140 kDa representing NPC1L1, and a nonspecific band may be observed at ~45 kDa depending on the lysate used.



Images

Western Blot: Niemann-Pick type C1 Like-1 Antibody [NB400-127] -Detection of NPC1L1 protein in lysate of human small intestine tissue using Niemann-Pick type C1 Like-1 antibody at 2ug/ml concentration.



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Immunocytochemistry/Immunofluorescence: Niemann-Pick type C1 Like-1 Antibody [NB400-127] - HepG2 cells were fixed in 4%

paraformaldehyde for 10 min and permeabilized in 0.05% Triton X-100 in PBS for 5 minutes. The cells were incubated with anti-Niemann-Pick type C1 Like-1 Antibody (NB400-127) at 2 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:500 dilution for 60 minutes. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 100X objective and digitally deconvolved.

Immunohistochemistry-Paraffin: Niemann-Pick type C1 Like-1 Antibody [NB400-127] - Analysis of a FFPE tissue section of mouse small intestine using 1:200 dilution of NPC1L1 antibody (NB400-127). The staining was developed using HRP labeled anti-rabbit secondary antibody and DAB reagent, and nuclei of cells were counter-stained with hematoxylin.





Publications

Lee C, Kwak SH, Han J et al. Repositioning of ezetimibe for the treatment of idiopathic pulmonary fibrosis. The European respiratory journal 2024-05-16 [PMID: 38359963]

Zhang Z, Qin S, Chen Y et al. Inhibition of NPC1L1 disrupts adaptive responses of drug-tolerant persister cells to chemotherapy EMBO molecular medicine 2022-01-13 [PMID: 35023619] (KD, Human)

Haghikia A, Zimmermann F, Schumann P Et al. Propionate attenuates atherosclerosis by immune-dependent regulation of intestinal cholesterol metabolism European heart journal 2021-10-01 [PMID: 34597388] (IHC-P)

Sugimoto S, Kobayashi E, Fujii M et al. An organoid-based organ-repurposing approach to treat short bowel syndrome Nature 2021-02-24 [PMID: 33627870]

Zimmermann F, Roessler J, Schmidt D et al. Impact of the Gut Microbiota on Atorvastatin Mediated Effects on Blood Lipids J Clin Med 2020-05-25 [PMID: 32466086] (IF/IHC, Mouse)

Nihei W, Nagafuku M, Hayamizu H et al. NPC1L1-dependent intestinal cholesterol absorption requires ganglioside GM3 in membrane microdomains. J. Lipid Res. 2018-09-21 [PMID: 30242108] (IF/IHC, Mouse)

Sachdev V, Leopold C, Bauer R et al. Novel role of a triglyceride-synthesizing enzyme: DGAT1 at the crossroad between triglyceride and cholesterol metabolism. Molecular and Cell Biology of Lipids. 2016-06-23 [PMID: 27344248]

Details:

This citation used the Alexa Fluor 488 version of this antibody.

Feng D, Zou J, Zhang S et al. Hypocholesterolemic Activity of Curcumin is Mediated by Down-Regulating the Expression of Niemann-Pick C1-Like 1 in Hamsters. J. Agric. Food Chem. 2016-12-21 [PMID: 28000447]

Kikuchi T, Orihara K, Oikawa F et al. Intestinal CREBH overexpression prevents high-cholesterol diet-induced hypercholesterolemia by reducing Npc1I1 expression. Mol Metab. 2016-11-01 [PMID: 27818935] (WB, Mouse)

Alvaro A, Rosales R, Masana L et al. Polyunsaturated fatty acids down-regulate in vitro expression of the key intestinal cholesterol absorption protein NPC1L1: no effect of monounsaturated nor saturated fatty acids. J Nutr Biochem 2010-06-01 [PMID: 19443194]

Skov M, Tonnesen CK, Hansen GH et al. Dietary cholesterol induces trafficking of intestinal Niemann-Pick Type C1 Like 1 from the brush border to endosomes. Am J Physiol Gastrointest Liver Physiol 2011-01-01 [PMID: 21051527] (IHC-P, EM, WB, Porcine)

Park Y, Carr TPet al. Unsaturated fatty acids and phytosterols regulate cholesterol transporter genes in Caco-2 and HepG2 cell lines. Nutr Res. 2013-02-01 [PMID: 23399666] (WB, Human)

More publications at http://www.novusbio.com/NB400-127





Procedures

Western Blot protocol for Niemann-Pick type C1 Like-1 Antibody (NB400-127)

Western Blot Protocol

1. Perform SDS-PAGE (3-8%) on samples to be analyzed, loading 50ug of total protein per lane.

2. Transfer proteins to Nitrocellulose according to the instructions provided by the manufacturer of the transfer apparatus.

3. Stain the blot using ponceau S for 1-2 minutes to access the transfer of proteins onto the nitrocellulose membrane. Rinse the blot in water to remove excess stain and mark the lane locations and locations of molecular weight markers using a pencil.

4. Rinse the blot in TBS for approximately 5 minutes.

5. Block the membrane using 5% non-fat dry milk in TBS for 1 hour at room temprature.

6. Dilute the rabbit anti-NPC1L1 primary antibody (NB 400-127) in 5% BSA and incubate overnight at 4 degreesCelcius.

7. Wash the membrane in water for 5 minutes and apply the diluted rabbit-IgG HRP-conjugated secondary antibody inblocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.

8. Wash the blot in TBS containing 0.05-0.1% Tween-20 for 10-20 minutes.

9. Wash the blot in type I water for an additional 10-20 minutes (this step can be repeated as required to reducebackground).

10. Apply the detection reagent of choice in accordance with the manufacturers instructions (Amersham's ECL is the standard reagent used at Novus Biologicals).

**Note: Tween-20 can be added to the blocking buffer at a final concentration of 0.05-0.2%, provided it does not interfere

with antibody-antigen binding.

Immunohistochemistry-Paraffin protocol for Niemann-Pick type C1 Like-1 Antibody (NB400-127)

Immunohistochemistry-paraffin embedded sections

Antigen Unmasking

- Bring slides to a boil in 10 mM sodium citrate buffer pH 6.0 then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench top for 30 minutes.

Staining

- Wash sections in dH2O three times for 5 minutes each.

- Wash section in wash buffer (1X PBS/0.1% Tween-20 (1X PBST)) for 5 minutes.

- Block each section with 100-400 ul blocking solution (1X PBST, 5% goat serum) for 1 hour at room temperature.

- Remove blocking solution and add 100-400 ul primary antibody diluted in 1X PBST, 5% goat serum to each section. Incubate overnight at 4C.

- Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.

- Add 100-400 ul biotinylated secondary antibody, diluted in 1X PBST, 5% goat serum. Incubate 30 minutes at room temperature.

- Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.

- Add 100-400 ul Striptavidin reagent to each section and incubate for 30 minutes at room temperature.

- Wash sections three times in wash buffer for 5 minutes each.
- Add 100-400 ul DAB substrate to each section and monitor staining closely.
- As soon as the sections develop, immerse slides in dH2O.
- Counterstain sections in hematoxylin.
- Wash sections in dH2O two times for 5 minutes each.
- Dehydrate sections.
- Mount coverslips.





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Products Related to NB400-127

NB400-127PEP	Niemann-Pick type C1 Like-1 Antibody Blocking Peptide
HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control

Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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