Product Datasheet

VPS34 Antibody NB110-87320SS

Unit Size: 0.025 ml

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.



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NB110-87320SS

VPS34 Antibody

| Product Information | |
|-----------------------------|--|
| Unit Size | 0.025 ml |
| Concentration | 0.2 mg/ml |
| Storage | Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles. |
| Clonality | Polyclonal |
| Preservative | 0.05% Sodium Azide |
| Isotype | IgG |
| Purity | Immunogen affinity purified |
| Buffer | PBS, 0.1% BSA, and 50% Glycerol |
| Target Molecular Weight | 100 kDa |
| Product Description | |
| Host | Rabbit |
| Gene ID | 5289 |
| Gene Symbol | PIK3C3 |
| Species | Human, Mouse, Rat, Porcine |
| Reactivity Notes | Immunogen displays the following percentage of sequence identity for non- tested species: Xenopus (87%). |
| Immunogen | Synthetic peptide made to an internal portion of the human VPS34 protein (within residues 700-850). [Swiss-Prot# Q8NEB9] |
| Product Application Details | |
| Applications | Western Blot, Simple Western, Immunocytochemistry/ Immunofluorescence, Knockdown Validated |
| Recommended Dilutions | Western Blot 1:1000, Simple Western 1:20, Immunocytochemistry/ Immunofluorescence 1:100, Knockdown Validated |
| Application Notes | This VPS34 antibody is useful for Immunocytochemistry/Immunofluorescence and Western blot, where a band is seen approx. 100 kDa. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. See <u>Simple Western Antibody Database</u> for Simple Western validation: Tested in HepG2 lysate 0.5 mg/mL, separated by Size, antibody dilution of 1:20, apparent MW was 102 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue. |

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Western Blot: VPS34 Antibody [NB110-87320] - Celastrol regulates autophagy- & mitophagy-related gene expressions in neurons. SH-SY5Y cells treated with 0.1–1 µM celastrol for 4 h. (A) Real-time quantitative PCR results show that 1 µM celastrol treatment for 4 h enhanced mRNA expressions of phagophore induction genes Beclin 1 & Ambra1, phagophore elongation genes Atg7 & Atg12, & vacuole formation genes LC3A & Atg4B. (B) Western blot results of SH-SY5Y treated with 100-500 nM celastrol show celastrol (500 nM) increased in protein expressions of Beclin 1 & Vps34 (phagophore induction), Atg7 (phagophore elongation), LC3-II (vacuole formation), DJ-1, & PINK1 (mitophagy). Celastrol (500 nM) suppressed LRRK2 expression. (C) MPTP (10 mg/kg/day for 3 days, i.p.) but not celastrol (3 mg/kg/day for 3 days, i.p.) caused dopaminergic nerve terminal degeneration (tyrosine hydroxylase) & mitophagy inactivation (DJ-1) & PINK1) in the striatum of mice as compared to saline control. Celastrol cotreatment with MPTP reversed it as compared to MPTP. Data are given as the mean ± SEM (n = 3–5). p-value was determined using the Kruskal–Wallis test followed by Dunn's multiple comparison post hoc test. * p < 0.05 compared with controls. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/31906147), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Yang D, Wu X, Wang W et al. Ciliary Type III Adenylyl Cyclase in the VMH Is Crucial for High-Fat Diet-Induced Obesity Mediated by Autophagy Advanced Science 2022-01-01 [PMID: 34783461] (Western Blot)

Meena N, Ralston E, Raben N, Puertollano R Enzyme Replacement Therapy Can Reverse Pathogenic Cascade in Pompe Disease Mol Ther Methods Clin Dev 2020-07-17 [PMID: 32671132] (FLOW, Human)

Rijal R, Cadena LA, Smith MR et al. Polyphosphate is an extracellular signal that can facilitate bacterial survival in eukaryotic cells Proceedings of the National Academy of Sciences of the United States of America 2020-12-15 [PMID: 33268492]

Lin M W, Lin C C et al. Celastrol Inhibits Dopaminergic Neuronal Death of Parkinson's Disease through Activating Mitophagy. Antioxidants (Basel) 2019-12-31 [PMID: 31906147] (WB, Mouse)

Campa CC, Margaria JP, Derle A et al. Rab11 activity and PtdIns(3)P turnover removes recycling cargo from endosomes Nat. Chem. Biol. 2018-06-18 [PMID: 29915378] (WB)

Brigger D, Schlafli AM, Garattini E, Tschan MP. Activation of RARa induces autophagy in SKBR3 breast cancer cells and depletion of key autophagy genes enhances ATRA toxicity. Cell Death Dis 2015-08-28 [PMID: 26313912] (WB, Human)

Huang Y, Hou JK, Chen TT et al. PML-RAR alpha enhances constitutive autophagic activity through inhibiting the Akt/mTOR pathway. Autophagy. 2011-10-01 [PMID: 21673516] (WB, Human)

Shi JM, Bai LL, Zhang DM et al. Saxifragifolin D induces the interplay between apoptosis and autophagy in breast cancer cells through ROS-dependent endoplasmic reticulum stress. Biochem Pharmacol 2013-01-21 [PMID: 23348250] (WB, Human)



Procedures

Serum protocol for VPS34 Antibody (NB110-87320)

VPS34 Antibody: Western Blot Protocol

1. Perform SDS-PAGE (4-12% MOPS) on samples to be analyzed, loading 40 ug of total protein per lane.

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

3. Rinse membrane with dH2O and then 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% BSA in TBS + Tween, 1 hour at RT.

6. Rinse the membrane in dH2O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.

7. Dilute the rabbit anti-VPS34 primary antibody (NB 110-87320) in blocking buffer and incubate 1 hour at room temperature.

8. Rinse the membrane in dH2O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.

9. Apply the diluted rabbit-IgG HRP-conjugated secondary antibody in blocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.

10. Wash the blot in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each (this step can be repeated as required to reduce background).

11. Apply the detection reagent of choice in accordance with the manufacturers instructions (Pierce ECL).

Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%, provided it does not interfere with antibody-antigen binding.

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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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