Product Datasheet

SMAD6 Antibody - BSA Free
NB100-56440

Unit Size: 0.1 mg

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

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Reviews: 2 Publications: 10

Protocols, Publications, Related Products, Reviews, Research Tools and Images at:
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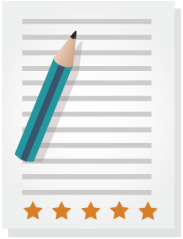
Updated 4/13/2025 v.20.1



NB100-56440

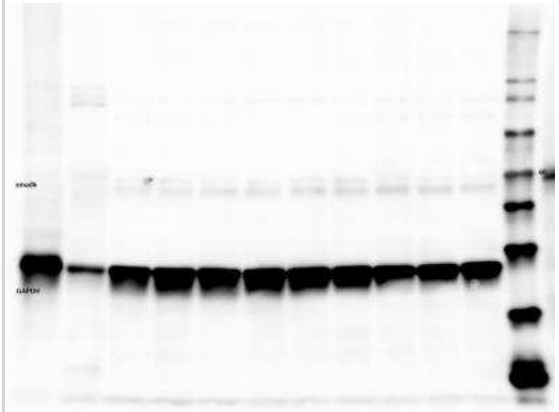
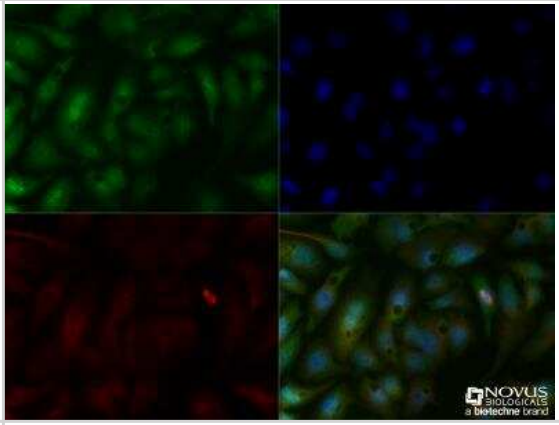
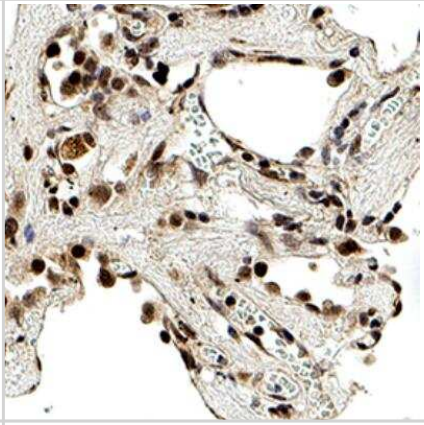
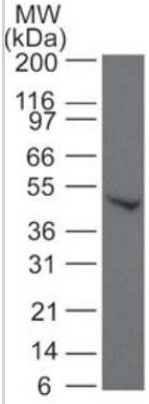
SMAD6 Antibody - BSA Free

Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.02% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS

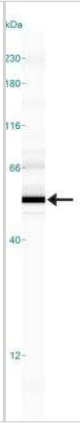
Product Description	
Host	Rabbit
Gene ID	4091
Gene Symbol	SMAD6
Species	Human, Mouse, Rat, Primate, Monkey, Sheep
Reactivity Notes	Cross reacts with New World Monkey. Predicted to react with Equine. Mouse reactivity reported from a customer review.
Immunogen	Synthetic peptide made to an internal portion of human SMAD6 (between amino acids 30-150) [UniProt O43541].

Product Application Details	
Applications	Western Blot, Simple Western, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 0.5 - 2 ug/mL. Use reported by customer review, Simple Western 1:50, Immunohistochemistry 5 - 10 ug/mL, Immunocytochemistry/ Immunofluorescence 1 - 5 ug/mL. Use reported in scientific literature (PMID 20032053), Immunohistochemistry-Paraffin 5 - 10 ug/mL. Use reported in scientific literature (PMID 20032053)
Application Notes	A 52 kDa band is observed. Use of this antibody in IHC-P and ICC-IF applications is cited by Hogg et al 2010 in Endocrinology 151:1247-56 (PMID: 20032053) and this publication has documented protocols for these applications. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. See Simple Western Antibody Database for Simple Western validation: Tested in HepG2 lysate 0.5 mg/mL, separated by Size, antibody dilution of 1:50, apparent MW was 54 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue.

Images

Western Blot: SMAD6 Antibody [NB100-56440] - Analysis in mouse adult lung tissue. WB image submitted by a verified customer review.	
Immunocytochemistry/Immunofluorescence: SMAD6 Antibody [NB100-56440] - HepG2 cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X TBS + 0.5% Triton X-100. The cells were incubated with anti-SMAD6 (NB100-56440) at a 1:100 dilution overnight at 4C and detected with an anti-rabbit DyLight 488 (Green) at a 1:500 dilution. Alpha tubulin was used as a co-stain at a 1:1000 dilution and detected with an anti-mouse DyLight 550 (Red) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.	
Immunohistochemistry-Paraffin: SMAD6 Antibody [NB100-56440] - SMAD6 was detected in immersion fixed paraffin-embedded sections of human lung using Rabbit Anti-Human SMAD6 polyclonal Antibody (Catalog # NB100-56440) at 5 ug/ml for 1 hour at room temperature followed by incubation with the Anti-Rabbit IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC003). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Nuclear staining was localized to the pneumocytes.	
Western Blot: SMAD6 Antibody [NB100-56440] - Analysis using SMAD6 antibody. Lysate from Jurkat cells probed with SMAD6 antibody at 2 ug/ml.	

Simple Western: SMAD6 Antibody [NB100-56440] - Simple Western lane view shows a specific band for SMAD6 in 0.5 mg/ml of HepG2 lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



Publications

Zagorka Vitic, Hazem Safory, Vukasin M Jovanovic, Yael Sarusi, Alexandra Stavsky, Joy Kahn, Alona Kuzmina, Lilah Toker, Daniel Gitler, Ran Taube, Roland H Friedel, Simone Engelender, Claude Brodski BMP5/7 protect dopaminergic neurons in an α-synuclein mouse model of Parkinson's disease Brain 2021-02-01 [PMID: 33253359]

Zuidhof H, Müller C, Kortman G et al. Oncogenic functions of the m6A demethylase FTO in breast cancer cells involving translational upregulation of C/EBP?-LIP bioRxiv 2023-09-23 (WB, Human)

Zhang X, Hu Z, Wang X Et Al. ANXA10 promotes melanoma metastasis by suppressing E3 ligase TRIM41-directed PKD1 degradation Cancer letters 2021-07-26 [PMID: 34324862]

Nakano Y, Hasegawa T, Kashino C et al. Aldosterone enhances progesterone biosynthesis regulated by bone morphogenetic protein in rat granulosa cells J. Steroid Biochem. Mol. Biol. 2020-08-21 [PMID: 32828828] (WB, Rat)

Yamaguchi H, Zhu J, Yu T et al. Low-level bisphenol A increases production of glial fibrillary acidic protein in differentiating astrocyte progenitor cells through excessive STAT3 and Smad1 activation. Toxicology. 2006-09-21 [PMID: 16860915]

Shen R, Chen M, Wang YJ et al. Smad6 interacts with Runx2 and mediates Smad ubiquitin regulatory factor 1-induced Runx2 degradation. J Biol Chem. 2006-02-10 [PMID: 16299379] (WB)

Details:
[WB, Fig 2f (COS monkey kidney cells, 2C12 mouse myoblast/osteoblast cells), WB, Fig 5a (Specificity of antibody validated by shRNA knockdown in 2T3 cells)]

Yoshiura S, Ohtsuka T, Takenaka Y et al. Ultradian oscillations of Stat, Smad, and Hes1 expression in response to serum. Proc Natl Acad Sci U S A. 2007-07-03 [PMID: 17592117] (WB, Mouse)

Details:
WB, Fig 3 (mouse C3HIOT1/2 fibroblasts).

Hogg K, Etherington SL, Young JM et al. Inhibitor of differentiation (Id) genes are expressed in the steroidogenic cells of the ovine ovary and are differentially regulated by members of the transforming growth factor-beta family. Endocrinology [PMID: 20032053] (IF/IHC, Sheep)

Details:
Used the Azide Free form of this antibody.

Wang Q, Wei X, Zhu T et al. Bone morphogenetic protein 2 activates Smad6 gene transcription through bone-specific transcription factor Runx2. J Biol Chem. 2007-04-06 [PMID: 17215250] (WB, Mouse)

Details:
WB, Fig. 1 (2C12 mouse myoblast/osteoblast cells).

Childs AJ, Kinnell HL, Collins CS et al. BMP signaling in the human fetal ovary is developmentally regulated and promotes primordial germ cell apoptosis. Stem Cells. 2010-08-01 [PMID: 20506112] (IHC-P, Human)

Details:
SMAD6 (IMG-555). IHC (Bouin's fixed, paraffin-embedded): Human fetal ovary, Fig 3A-3D.

Procedures

Western Blot Protocol for SMAD6 Antibody (NB100-56440)
Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 10-25 ug of total protein per lane.
2. Transfer proteins to PVDF membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
3. Stain the membrane with Ponceau S (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
4. Rinse the blot TBS -0.05% Tween 20 (TBST).
5. Block the membrane in 5% Non-fat milk in TBST (blocking buffer) for at least 1 hour.
6. Wash the membrane in TBST three times for 10 minutes each.
7. Dilute primary antibody in blocking buffer and incubate overnight at 4C with gentle rocking.
8. Wash the membrane in TBST three times for 10 minutes each.
9. Incubate the membrane in diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturer's instructions) for 1 hour at room temperature.
10. Wash the blot in TBST three 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 manufacturer's instructions.

Immunohistochemistry-Paraffin Protocol for SMAD6 Antibody (NB100-56440)
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 (keep slides in the sodium citrate buffer at all times).

Staining:
1. Wash sections in deionized water three times for 5 minutes each.
2. Wash sections in PBS for 5 minutes.
3. Block each section with 100-400 ul blocking solution (1% BSA in PBS) for 1 hour at room temperature.
4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 C.
5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
6. Add 100-400 ul HRP polymer conjugated secondary antibody. Incubate 30 minutes at room temperature.
7. Wash sections three times in wash buffer for 5 minutes each.
8. Add 100-400 ul DAB substrate to each section and monitor staining closely.
9. As soon as the sections develop, immerse slides in deionized water.
10. Counterstain sections in hematoxylin.
11. Wash sections in deionized water two times for 5 minutes each.
12. Dehydrate sections.
13. Mount coverslips.

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Products Related to NB100-56440

NB800-PC2	Jurkat Whole Cell Lysate
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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