Product Datasheet

Smad3 [p Ser423, p Ser425] Antibody - BSA Free
NBP1-77836

Unit Size: 0.1 mg

Store at -20C short term. Aliquot and store at -80C long term. Avoid freeze-thaw cycles.

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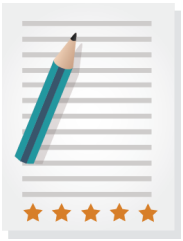
Updated 2/21/2025 v.20.1



NBP1-77836

Smad3 [p Ser423, p Ser425] Antibody - BSA Free

Product Information

Unit Size	0.1 mg
Concentration	Please see the vial label for concentration. If unlisted please contact technical services.
Storage	Store at -20C short term. Aliquot and store at -80C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.01% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

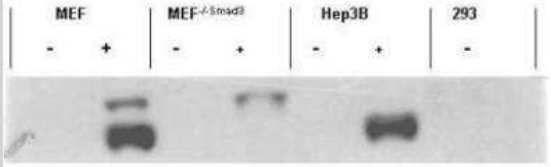
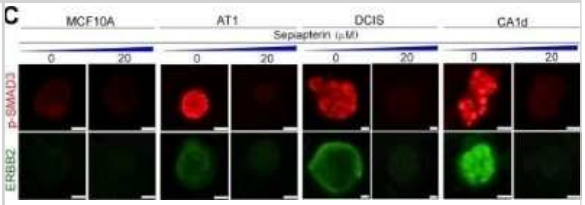
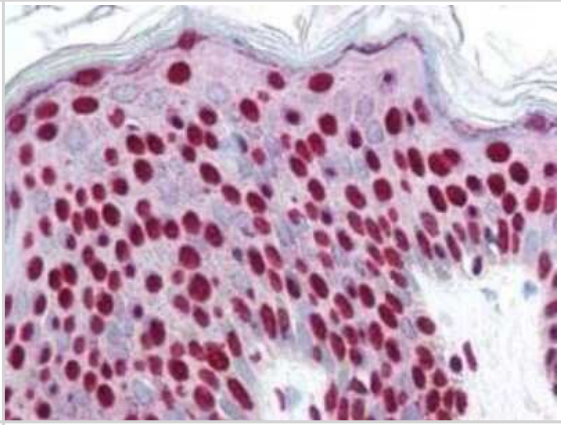
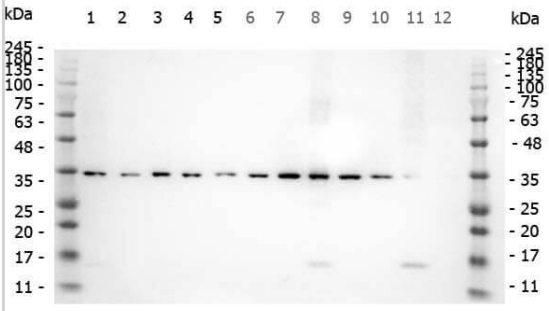
Product Description

Description	This affinity-purified antibody is directed against the phosphorylated form of human Smad3 protein at the [p Ser423] and [p Ser425] residues. The product was affinity purified from monospecific antiserum by immunoaffinity purification. Antiserum was first purified against the phosphorylated form of the immunizing peptide. The resultant affinity purified antibody was then cross adsorbed against the non-phosphorylated form of the immunizing peptide Store vial at -20C prior to opening. Aliquot contents and freeze at -20C 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 4C as an undiluted liquid. Dilute only prior to immediate use.
Host	Rabbit
Gene ID	4088
Gene Symbol	SMAD3
Species	Human, Mouse
Reactivity Notes	A BLAST analysis was used to suggest cross reactivity with Smad3 from Xenopus laevis, Xenopus tropicalis, zebrafish, swine, bovine and chicken based on 100% sequence homology with the immunogen. Reactivity against homologues from other sources is not known
Specificity/Sensitivity	This affinity-purified antibody is directed against the phosphorylated form of human Smad3 protein at the pS423 and pS425 residues. The resultant affinity purified antibody was then cross adsorbed against the non-phosphorylated form of the immunizing peptide. Reactivity occurs against human Smad3 pS423 and pS425 protein and the antibody is specific for the phosphorylated form of the protein. Reactivity with non-phosphorylated human Smad3 is minimal by ELISA and western blot. Expect reactivity against phosphorylated Smad1 and Smad5. Negligible reactivity is seen against other phosphorylated Smad family members. A BLAST analysis was used to suggest cross reactivity with Smad3 from human, Xenopus laevis, Xenopus tropicalis, zebrafish, rat, mouse, swine, bovine and chicken based on 100% sequence homology with the immunogen. Reactivity against homologues from other sources is not known.
Immunogen	Smad3 [p Ser423, p Ser425] Antibody was prepared from whole rabbit serum produced by repeated immunizations with a dual phosphorylated synthetic peptide corresponding to a c-terminal region with Serine 423 and Serine 425 of human Smad3 protein. (Uniprot: P84022)

Product Application Details

Applications	Western Blot, ELISA, Flow Cytometry, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Single Cell Western
Recommended Dilutions	Western Blot 1:2000-1:20000, Flow Cytometry 1:10-1:1000, ELISA 1:15000-1:30000, Immunohistochemistry 1:500, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry-Paraffin 1:10-1:500, Single Cell Western 100 ug/ml
Application Notes	This affinity purified antibody has been tested for use in ELISA, immunohistochemistry, and western blot. Specific conditions for reactivity should be optimized by the end user. Expect a band approximately 48 kDa in size corresponding to phosphorylated Smad3 protein by western blotting in the appropriate stimulated tissue or cell lysate or extract. Less than 0.2% reactivity is observed against the non-phosphorylated form of the immunizing peptide. This antibody is phospho specific for dual phosphorylated pS423 and pS425 of Smad3. Stimulation with 2 ng/ml TGF-beta for 1 hour is suggested.

Images

Western Blot: Smad3 [p Ser423, p Ser425] Antibody [NBP1-77836] - Shows detection of endogenous Smad3 in stimulated cell lysates. Lysates were prepared from control cells (- lanes), or cells stimulated with 2 ng/ml TGF (+lanes) for 1 hour. This reagent recognizes phosphorylated Smad3 and has negligible reactivity against non-phosphorylated Smad3 protein. Personal Communication. YIgG Zhang, NIH, CCR, Bethesda, MD.	
Immunocytochemistry/Immunofluorescence: Smad3 [p Ser423, p Ser425] Antibody [NBP1-77836] - BH4 precursor, sepiapterin, normalizes NO level and ameliorates malignant phenotype of precancerous and cancerous cells. Representative images of the progression series cultivated in 3D ECM with or without sepiapterin (20 uM) and stained for phospho-SMAD3 or ERBB2. Nuclei were counterstained with DAPI (blue). Scale bars: 20 um. (Bottom) Quantification of phospho-SMAD3 (left) and ERBB2 levels (right) of the progression series. Error bars: mean +/- STDEV. *p < 0.05; **p < 0.01 and ***p < 0.001. Note that the effects of sepiapterin are only discernible in precancerous and cancerous cells, but not in non-malignant cells. (See the results of different concentrations of sepiapterin in Supplementary Fig. 8). Image collected and cropped by CiteAb from the following publication (www.nature.com/articles/s41598-019-43239-x) licensed under a CC-BY license.	
Immunohistochemistry: Smad3 [p Ser423, p Ser425] Antibody [NBP1-77836] - Used at 2.5 ug/ml to detect signal in a variety of tissues including multi-human, multi-brain and multi-cancer slides. This image shows strong nuclear staining in the majority of epidermal keratinocytes at 40X. Tissue was formalin-fixed and paraffin embedded. The image shows localization of the antibody as the precipitated red signal, with a hematoxylin purple nuclear counterstain. Personal Communication, Tina Roush, LifeSpanBiosciences, Seattle, WA.	
Western Blot of Rabbit anti-SMAD3 pS423 pS425 antibody. Marker: Opal Pre-stained ladder	

Publications

Van Krieken R, Marway M, Parthasarathy P, Mehta N et Al. Inhibition of SREBP With Fatostatin Does Not Attenuate Early Diabetic Nephropathy in Male Mice Endocrinology 2018-02-09 [PMID: 29420703]

Soomro A, Khajehei M, Li R et al. A therapeutic target for CKD: activin A facilitates TGF?1 profibrotic signaling Cellular & Molecular Biology Letters 2023-01-30 [PMID: 36717814] (ELISA, Western Blot)

Letson J, Furuta S. Reduced S-nitrosylation of TGF?1 elevates its binding affinity towards the receptor and promotes fibrogenic signaling in the breast bioRxiv 2023-09-12 [PMID: 37745487] (Western Blot, Immunocytochemistry/ Immunofluorescence)

Smith SS, Chu D, Qu T et al. Species-specific sensitivity to TGF? signaling and changes to the Mmp13 promoter underlie avian jaw development and evolution eLife 2022-06-06 [PMID: 35666955] (WB)

Cuevas RA, Wong R, Joolharzadeh P et al. Ecto-5'-Nucleotidase (Nt5e/CD73)-Mediated Adenosine Signaling Attenuates TGFb-2 Induced Elastin and Cellular Contraction American journal of physiology. Cell physiology 2022-12 -12 [PMID: 36503240] (Western Blot, Mouse)

Mukhi D, Kolligundla LP, Maruvada S et al. Growth hormone induces transforming growth factor-β1 in podocytes: Implications in podocytopathy and proteinuria Biochimica et biophysica acta. Molecular cell research 2022-11-15 [PMID: 36400249] (WB, Human)

Bourdon R Regulation of epinephrine biosynthesis by intermittent and continuous hypoxia Nat Commun 2020-02-07 [PMID: 32024825] (IF/IHC, Mouse)

Ren G, Zheng X, et al. Reduced Basal Nitric Oxide Production Induces Precancerous Mammary Lesions via ERBB2 and TGF beta. Sci Rep 2019-04-30 [PMID: 31040372] (ICC/IF, Human)

Zheng X, Fernando V, Sharma V et al. Correction of Arginine Metabolism with Sepiapterin-the Precursor of Nitric Oxide Synthase Cofactor BH4-Induces Immunostimulatory-shift of Breast Cancer Biochem. Pharmacol. 2020-02-26 [PMID: 32112882] (WB, Human)

Huber AK, Sorkin M, Hwang C, et al. Regulation of heterotopic ossification through local inflammatory monocytes in a mouse model of aberrant wound healing bioRxiv (ICC/IF, Mouse)

Huber AK, Sorkin M, Hwang C, et al. Regulation of heterotopic ossification through local inflammatory monocytes in a mouse model of aberrant wound healing bioRxiv (ICC/IF, Mouse)

Rogers KA, Moreno SE, Smith LA et al. Differences in the timing and magnitude of Pkd1 gene deletion determine the severity of polycystic kidney disease in an orthologous mouse model of ADPKD Physiol Rep 2016-06-01 [PMID: 27356569]

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NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control
NBP1-30284	Recombinant Human Smad3 His Protein

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