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Anti-Phospho-SMAD3 S423/S425 Antibody

Catalog Number: P00059

About SMAD3

This antibody is suitable for Cancer, Immunology and Nuclear Signaling research. Smad3 (also known as Mothers against decapentaplegic homolog 3 Mothers against DPP homolog 3, Mad3, hMAD-3, JV15-2 or hSMAD3) is a transcriptional modulator activated by TGF-beta (transforming growth factor) and activin type 1 receptor kinase. These activators exert diverse effects on a wide array of cellular processes. The Smad proteins mediate much of the signaling responses induced by the TGF-b superfamily. Briefly, activated type I receptor phosphorylates receptor-activated Smads (R-Smads) at their c-terminal two extreme serines in the SSXS motif, e.g. Smad2 and Smad3 proteins in the TGF-b pathway, or Smad1, Smad5 or Smad8 in the BMP pathway. Then the phosphorylated R-Smad translocated into nucleus, where they regulate transcription of target genes. Based on microarray and animal model experiments, Smad3 accounts for at least 80% of all TGF-b-mediated response.

Overview

Product Name	Anti-Phospho-SMAD3 S423/S425 Antibody
Reactive Species	Human
Description	Boster Bio Anti-Phospho-SMAD3 S423/S425 Antibody (Catalog # P00059). Tested in ELISA, IHC, WB applications. This antibody reacts with Human.
Application	ELISA, IHC, WB
Clonality	Polyclonal
Formulation	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2, 0.01% (w/v) Sodium Azide
Storage Instructions	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 date is one (1) year from date of opening. (Ship on dry ice.)
Host	Rabbit
Uniprot ID	P84022

Technical Details

Immunogen	Anti-SMAD3 pS423pS425 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.
Predicted Reactive Species	Bovine, Chicken, Xenopus Laevis, Xenopus Tropicalis, Zebrafish
Cross Reactivity	No cross reactivity with other proteins.
Isotype	IgG

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Form	Liquid (sterile filtered)
Concentration	1.16 mg/ml by UV absorbance at 280 nm
Purification	This affinity-purified antibody is directed against the phosphorylated form of human Smad3 protein at the pS423 and pS425 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. 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.
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit. If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples. Some PubMed article(s) citing the expression level of this target are as follows: Boster Bio's internal QC testing used: ELISA: 1:15,000 - 1:30,000 Flow Cytometry: User optimized IHC: 1:500 WB: 1:2,000 - 1:20,000

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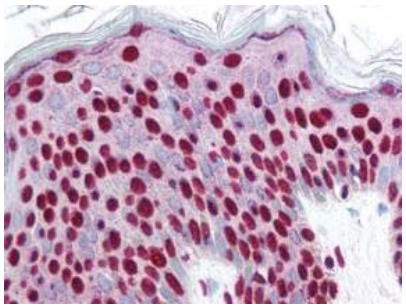
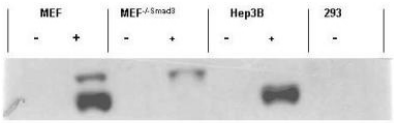
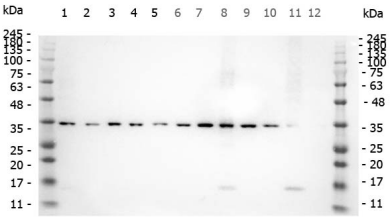
Anti-Phospho-SMAD3 S423/S425 Antibody (P00059) Images



SMAD3 was detected in paraffin-embedded sections of epidermal keratinocytes at 40X using rabbit anti-SMAD3 phospho S423/phospho S425 Antigen Affinity purified polyclonal antibody (Catalog # P00059) at 2.5 ug/mL. The image shows localization of the antibody as the precipitated red signal, with a hematoxylin purple nuclear counterstain. Personal Communi-cation, Tina Roush, LifeSpanBiosciences, Seattle, WA.



Western blot analysis of endogenous SMAD3 expression in stimulated cell lysates. Lysates were prepared from control cells (-lanes), or cells stimulated with 2 ng/ml TGF for 1 hour. Phosphorylated SMAD3 was detected using rabbit anti-SMAD3 phospho S423/phospho S425 Antigen Affinity purified polyclonal antibody (Catalog # P00059). Personal Communication. Ying Zhang, NIH, CCR, Bethesda, MD.



Western blot analysis of SMAD3 expression in HEK293 lysate (lane 1), HeLa Lysate (lane 2), MCF-7 Lysate (lane 3), Jurkat Lysate (lane 4), A431 Lysate (lane 5), A549 Lysate (lane 6), LNCap Lysate (lane 7), MOLT-4 Lysate (lane 8), Ramos Lysate (lane 9), Raji Lsyate (lane 10), A-172 Lysate (lane 11) and NIH/3T3 Lysate (lane 12). Phosphorylated SMAD3 at 35KD was detected using rabbit anti-SMAD3 phospho S423/phospho S425 Antigen Affinity purified polyclonal antibody (Catalog # P00059) at 1ug/mL. The blot was developed using chemiluminescence (ECL) method (Catalog # EK1002).

4 Publications Citing This Product

1. PubMed ID: 34093874, Wang Q,Liu J,Hu Y,Pan T,Xu Y,Yu J,Xiong W,Zhou Q,Wang Y.Local administration of liposomal-based Srpx2 gene therapy reverses pulmonary fibrosis by blockading fibroblast-to-myofibroblast transition.Theranostics.2021 May 13;11(14):7110-7125.doi:10.7150/thno.61085. PMID:34093874;PMCID:PMC8171094.

2. PubMed ID: 32826874, Hong L,Li F,Tang C,Li L,Sun L,Li X,Zhu L.Semaphorin 7A promotes endothelial to mesenchymal transition through ATF3 mediated TGF-beta2/Smad signaling.Cell Death Dis.2020 Aug 10;11(8):695.doi:10.1038/s41419-020-02818-x.PMID:32826874;PMCID:PMC7442651.

3. PubMed ID: 25820389, Xu T, Ni Mm, Huang C, Meng Xm, He Yh, Zhang L, Li J. Inflammation. 2015 Oct;38(5):1794-804. Doi: 10.1007/S10753-015-0157-6. Nlrc5 Mediates Il-6 And Il-1?? Secretion In Lx-2 Cells And Modulated By The Nf-??b/Smad3 Pathway.

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