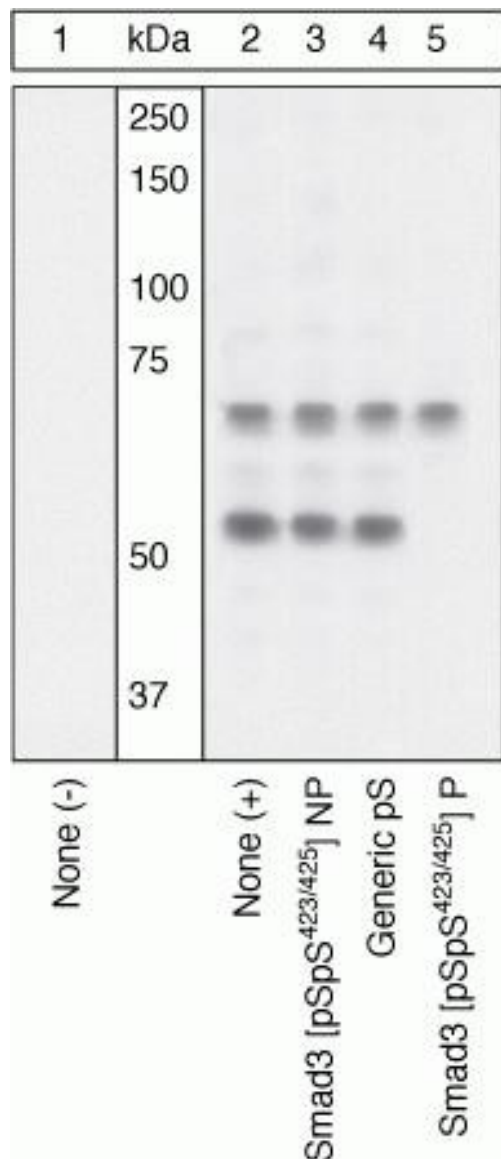


# Phospho-SMAD3 (Ser423, Ser425) Polyclonal Antibody

Product Details	
Size	100 µL
Species Reactivity	Human, Mouse
Published Species	Rat, Non-human primate, Hamster, Human, Mouse
Host/Isotype	Rabbit / IgG
Class	Polyclonal
Type	Antibody
Conjugate	Unconjugated
Immunogen	The antiserum was produced against a chemically synthesized phosphopeptide derived from a region of human Smad3 that contains serine 423 and serine 425. This sequence is conserved in mouse.
Form	Liquid
Purification	Antigen affinity chromatography
Storage buffer	Dulbecco's PBS, pH 7.3, with 1mg/mL BSA
Contains	0.05% sodium azide
Storage conditions	-20°C
RRID	AB_2533615

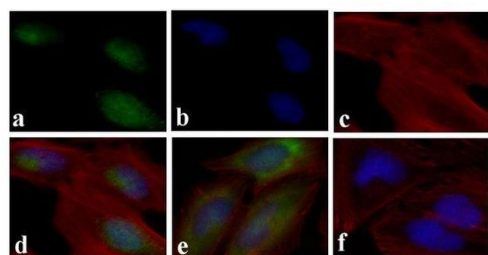
Applications	Tested Dilution	Publications
Western Blot (WB)	1:1,000	5 Publications
Immunohistochemistry (IHC)	-	4 Publications
Immunocytochemistry (ICC/IF)	1:250	2 Publications

## Product Images For Phospho-SMAD3 (Ser423, Ser425) Polyclonal Antibody



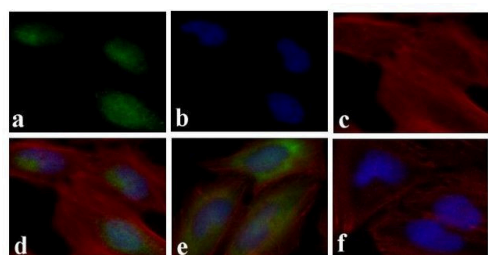
### Phospho-SMAD3 (Ser423, Ser425) Antibody (44-246G) in WB

Peptide Competition. Extracts of NMuMG cells unstimulated (1) or stimulated with 8 ng/mL TGF-beta for 20 minutes (2-5) were resolved by SDS-PAGE on a 10% Tris-glycine gel and transferred to PVDF. The membrane was blocked with a 5% BSA-TBST buffer for one hour at room temperature, then incubated with Smad3 (pSpS423/425) antibody for two hours at room temperature in a 3% BSA-TBST buffer, following prior incubation with: no peptide (1, 2), the non-phosphorylated peptide corresponding to the phosphopeptide immunogen (3), a generic phosphoserine-containing peptide (4), or the phosphopeptide immunogen (5). After washing, the membrane was incubated with goat F (ab')<sub>2</sub> anti-rabbit IgG HRP conjugate (Product # ALI4404) and signals were detected using the Pierce SuperSignal™ method. The data show that only the phosphopeptide corresponding to Smad3 (pSpS423/425) completely blocks the antibody signal, demonstrating the specificity of the antibody. The data also show upregulation of Smad3 (pSpS423/425) by stimulation with TGF-beta in this testing system. Note: The upper band is believed to be Smad3 in complex with Smad4.



### Phospho-SMAD3 (Ser423, Ser425) Antibody (44-246G)

Detection of altered subcellular localization of the target protein by cell treatment demonstrates antibody specificity. Immunofluorescence analysis of Phospho-SMAD3 (Ser423, Ser425) using Phospho-SMAD3 (Ser423, Ser425) Rabbit Polyclonal Antibody (Product # 44-246G) shows translocation of SMAD3 from cytoplasm to nucleus of HeLa cells upon treatment with TGF-beta. {TM}



### Phospho-SMAD3 (Ser423, Ser425) Antibody (44-246G) in ICC/IF

Immunofluorescence analysis of SMAD3 (pSpS423/425) was done on 70% confluent log phase HeLa cells treated with TGF-beta (20 ng/mL for 15 min) cells. The cells were fixed with 4% paraformaldehyde for 15 minutes, permeabilized with 0.25% Triton™ X-100 for 10 minutes, and blocked with 5% BSA for 1 hour at room temperature. The cells were labeled with SMAD3 (pSpS423/425) Rabbit polyclonal Antibody (Product # 44-246G) at 1:250 dilution in 1% BSA and incubated for 3 hours at room temperature and then labeled with Alexa Fluor 488 Goat Anti-Rabbit IgG Secondary Antibody (Product # A-11008) at a dilution of 1:400 for 30 minutes at room temperature (Panel a: green). Nuclei (Panel b: blue) were stained with SlowFade® Gold Antifade Mountant DAPI (Product # S36938). F-actin (Panel c: red) was stained with Alexa Fluor 594 Phalloidin (Product # A12381). Panel d is a merged image showing nuclear translocation of SMAD3 (pSpS423/425). Panel e cytoplasmic localization. Panel f shows no primary antibody control. The images were captured at 20X magnification.

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11 References

Western Blot (5)

<p>Molecular medicine reports</p> <p><b>Betulinic acid prevents high glucoseinduced expression of extracellular matrix protein in cardiac fibroblasts by inhibiting the TGF1/Smad signaling pathway.</b></p> <p>"44-246G was used in Western Blotting to study the effect of betulinic acid on high glucose induced cardiac fibroblasts and study the mechanism underlying this effect."</p> <p>Authors: Jiang L,Chen FX,Zang ST,Yang QF</p>	<p>Year 2017</p> <p>Species Rat</p> <p>Dilution 1:3,000</p>
<p>Scientific reports</p> <p><b>Restenosis Inhibition and Re-differentiation of TGF/Smad3-activated Smooth Muscle Cells by Resveratrol.</b></p> <p>"44246G was used in western blot to study the effects of periadventitial delivery of resveratrol on three major pro-restenotic pathologies"</p> <p>Authors: Zhu Y,Takayama T,Wang B,Kent A,Zhang M,Binder BY,Urabe G,Shi Y,DiRenzo D,Goel SA,Zhou Y,Little C,Roenneburg DA,Shi XD,Li L,Murphy WL,Kent KC,Ke J,Guo LW</p>	<p>Year 2017</p> <p>Species Human</p> <p>Dilution 1:1000</p>

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Immunohistochemistry (4)

<p>Scientific reports</p> <p><b>SMAD3 contributes to ascending aortic dilatation independent of transforming growth factor-beta in bicuspid and unicuspid aortic valve disease.</b></p> <p>"Published figure using Phospho-SMAD3 (Ser423, Ser425) polyclonal antibody (Product # 44-246G) in Immunohistochemistry"</p> <p>Authors: Balint B,Federspiel J,Kollmann C,Teping P,Schwab T,Schäfers HJ</p>	<p>Year 2022</p>
<p>Marine drugs</p> <p><b>Echinochrome A Treatment Alleviates Fibrosis and Inflammation in Bleomycin-Induced Scleroderma.</b></p> <p>"Published figure using Phospho-SMAD3 (Ser423, Ser425) polyclonal antibody (Product # 44-246G) in Immunohistochemistry"</p> <p>Authors: Park GT,Yoon JW,Yoo SB,Song YC,Song P,Kim HK,Han J,Bae SJ,Ha KT,Mishchenko NP,Fedoreyev SA, Stonik VA,Kim MB,Kim JH</p>	<p>Year 2021</p>

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ICC/IF (2)

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