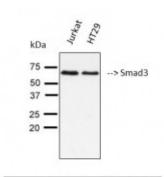


## Anti-Smad3 antibody



Western Blot (WB) analysis of Jurkat and HT29 cell lysate using Smad3 antibody (STJ95699).



**Description** Smad3 is a protein encoded by the SMAD3 gene which is approximately

48 kDa. Smad3 is localised to the cytoplasm and nucleus. It is involved in SMAD2/3 MH2 domain mutants in cancer, the cell cycle and toll-like receptor signalling pathway. It an intracellular signal transducer and transcriptional modulator activated by TGF-beta and activin type 1 receptor kinases. It binds the TRE element in the promoter region of many genes that are regulated by TGF-beta and, on formation of the SMAD3/SMAD4 complex, activates transcription. Smad3 is expressed in the nervous system, pancreas, lung, kidney and intestine. Mutations in the SMAD3 gene may result in colorectal cancer and Loeys-Dietz syndrome. STJ95699 was affinity-purified from rabbit antiserum by affinity-

chromatography using epitope-specific immunogen. This polyclonal

antibody detects endogenous levels of Smad3 protein.

Model STJ95699

Host Rabbit

Reactivity Human, Mouse, Rat

**Applications** ELISA, IHC, WB

**Immunogen** Synthesized peptide derived from human Smad3 around the non-

phosphorylation site of T179.

120-200 aa **Immunogen Region** 

**Gene ID** 4088

Gene Symbol SMAD3

WB 1:500-1:2000IHC 1:100-1:300ELISA 1:10000 **Dilution range** 

**Specificity** Smad3 Polyclonal Antibody detects endogenous levels of Smad3 protein.

**Purification** The antibody was affinity-purified from rabbit antiserum by affinity-

chromatography using epitope-specific immunogen.

**Note** For Research Use Only (RUO).

Protein Name Mothers against decapentaplegic homolog 3 MAD homolog 3 Mad3 Mothers

against DPP homolog 3 hMAD-3 JV15-2 SMAD family member 3 SMAD 3

Smad3 hSMAD3

Molecular Weight 50 kDa

**Clonality** Polyclonal

**Conjugation** Unconjugated

**Isotype** IgG

**Formulation** Liquid in PBS containing 50% glycerol, 0.5% BSA and 0.02% sodium azide.

**Concentration** 1 mg/ml

**Storage Instruction** Store at -20°C, and avoid repeat freeze-thaw cycles.

Database Links <u>HGNC:6769OMIM:114500</u>

Alternative Names Mothers against decapentaplegic homolog 3 MAD homolog 3 Mad3 Mothers

against DPP homolog 3 hMAD-3 JV15-2 SMAD family member 3 SMAD 3

Smad3 hSMAD3

**Function** Receptor-regulated SMAD (R-SMAD) that is an intracellular signal

transducer and transcriptional modulator activated by TGF-beta (transforming growth factor) and activin type 1 receptor kinases. Binds the TRE element in the promoter region of many genes that are regulated by TGF-beta and, on formation of the SMAD3/SMAD4 complex, activates transcription. Also can form a SMAD3/SMAD4/JUN/FOS complex at the AP-1/SMAD site to regulate TGF-beta-mediated transcription. Has an inhibitory effect on wound healing probably by modulating both growth and migration of primary keratinocytes and by altering the TGF-mediated chemotaxis of monocytes. This effect on wound healing appears to be hormone-sensitive. Regulator of chondrogenesis and osteogenesis and inhibits early healing of bone fractures. Positively regulates PDPK1 kinase activity by stimulating its dissociation from the 14-3-3 protein YWHAQ which acts as a negative regulator.

Sequence and Domain Family The MH1 domain is required for DNA binding. Also binds zinc ions which

are necessary for the DNA binding.; The MH2 domain is required for both homomeric and heteromeric interactions and for transcriptional regulation. Sufficient for nuclear import.; The linker region is required for the TGFbeta-mediated transcriptional activity and acts synergistically with the MH2

domain.

Cellular Localization Cytoplasm Nucleus. Cytoplasmic and nuclear in the absence of TGF-beta. On

TGF-beta stimulation, migrates to the nucleus when complexed with SMAD4

. Through the action of the phosphatase PPM1A, released from the

SMAD2/SMAD4 complex, and exported out of the nucleus by interaction with RANBP1 . Co-localizes with LEMD3 at the nucleus inner membrane . MAPK-mediated phosphorylation appears to have no effect on nuclear import

. PDPK1 prevents its nuclear translocation in response to TGF-beta .

## Post-translational Modifications

Phosphorylated on serine and threonine residues. Enhanced phosphorylation in the linker region on Thr-179, Ser-204 and Ser-208 on EGF and TGF-beta treatment. Ser-208 is the main site of MAPK-mediated phosphorylation. CDK-mediated phosphorylation occurs in a cell-cycle dependent manner and inhibits both the transcriptional activity and antiproliferative functions of SMAD3. This phosphorylation is inhibited by flavopiridol. Maximum phosphorylation at the G(1)/S junction. Also phosphorylated on serine residues in the C-terminal SXS motif by TGFBR1 and ACVR1. TGFBR1mediated phosphorylation at these C-terminal sites is required for interaction with SMAD4, nuclear location and transactivational activity, and appears to be a prerequisite for the TGF-beta mediated phosphorylation in the linker region. Dephosphorylated in the C-terminal SXS motif by PPM1A. This dephosphorylation disrupts the interaction with SMAD4, promotes nuclear export and terminates TGF-beta-mediated signaling. Phosphorylation at Ser-418 by CSNK1G2/CK1 promotes ligand-dependent ubiquitination and subsequent proteasome degradation, thus inhibiting SMAD3-mediated TGFbeta responses. Phosphorylated by PDPK1. Acetylation in the nucleus by EP300 in the MH2 domain regulates positively its transcriptional activity and is enhanced by TGF-beta. Poly-ADP-ribosylated by PARP1 and PARP2. ADP-ribosylation negatively regulates SMAD3 transcriptional responses during the course of TGF-beta signaling. Ubiquitinated. Monoubiquitinated, leading to prevent DNA-binding. Deubiquitination by USP15 alleviates inhibition and promotes activation of TGF-beta target genes. Ubiquitinated by RNF111, leading to its degradation: only SMAD3 proteins that are 'in use' are targeted by RNF111, RNF111 playing a key role in activating SMAD3 and regulating its turnover. Undergoes STUB1-mediated ubiquitination and degradation.

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