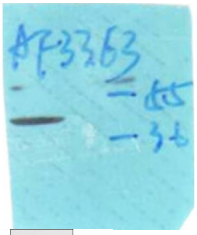
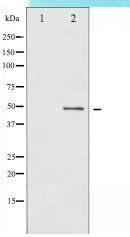
Phospho-Smad3 (Thr179) Ab

Cat.#: AF3363	Concn.: 1mg/ml	Mol.Wt.: 50kDa
Size: 100ul,200ul	Source: Rabbit	Clonality: Polyclonal

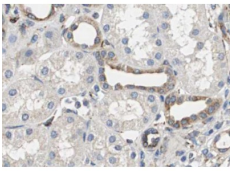
Application:	WB 1:500-1:2000 IHC 1:50-1:200, IF/ICC 1:100-1:500
Reactivity:	Human,Mouse,Rat
Purification:	The Ab is from purified rabbit serum by affinity purification via sequential chromatography on phospho- and non-phospho-peptide affinity columns.
Specificity:	Phospho-Smad3 (Thr179) Ab detects endogenous levels of Smad3 only when phosphorylated at Threonine 179.
Immunogen:	A synthesized peptide derived from human Smad3 around the phosphorylation site of Threonine 179.
Uniprot:	P84022
Description:	Smad3 transcription factor phosphorylated and activated by TGF-beta-type receptors. A receptor-regulated Smad (R-smad). Binds directly to consensus DNA-binding elements in the promoters of target genes. In mouse required for establishemnt of the mucosal immune response and proper development of skeleton.
Subcellular Location:	Cytoplasm. Nucleus. Cytoplasmic and nuclear in the absence of TGF-beta. On TGF-beta stimulation, migrates to the nucleus when complexed with SMAD4 (PubMed:15799969). Through the action of the phosphatase PPM1A, released from the SMAD2/SMAD4 complex, and exported out of the nucleus by interaction with RANBP1 (PubMed:16751101, PubMed:19289081). Co-localizes with LEMD3 at the nucleus inner membrane (PubMed:15601644). MAPK-mediated phosphorylation appears to have no effect on nuclear import (PubMed:19218245). PDPK1 prevents its nuclear translocation in response to TGF-beta (PubMed:17327236).
Similarity:	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.Belongs to the dwarfin/SMAD family.
Storage Condition and Buffer:	Rabbit IgG in phosphate buffered saline , pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol.Store at -20 °C.Stable for 12 months from date of receipt.



Western blot analysis of Phospho-Smad3 (Thr179) Ab expression in TNF treated Hela cells lysates.The lane on the right is treated with the antigen-specific peptide.



Western blot analysis of Smad3 phosphorylation expression in TNF treated HeLa whole cell lysates,The lane on the left is treated with the antigen-specific peptide.



AF3363 at 1/100 staining human kidney tissue sections by IHC-P. The tissue was formaldehyde fixed and a heat mediated antigen retrieval step in citrate buffer was performed. The tissue was then blocked and incubated with the Ab for 1.5 hours at 22°C. An HRP conjugated goat anti-rabbit Ab was used as the secondary.

AF3363 staining NIH-3T3 by IF/ICC. The sample were fixed with PFA and permeabilized in 0.1% Triton X-100,then blocked in 10% serum for 45 minutes at 25°C. The primary Ab was diluted at 1/200 and incubated with the sample for 1 hour at 37°C. An Alexa Fluor 594 conjugated goat anti-rabbit IgG (H+L) Ab, diluted at 1/600, was used as the secondary Ab.

IMPORTANT: For western blot, incubate membrane with diluted primary Ab in 5% w/v milk , 1X TBS, 0.1% Tween®20 at 4°C with gentle shaking, overnight.