Anti-Phospho-Smad2 (S255) Antibody [JF0882] ET1702-34

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IP
Molecular Wt:	Predicted band size: 52 kDa
Clone number:	JF0882

Description: Smad proteins, the mammalian homologs of the Drosophila mothers against decapentaplegic (Mad), have been implicated as downstream effectors of TGFβ/BMP signaling. Smad1 (also designated Madr1 or JV4-1) and Smad5 are effectors of BMP-2 and BMP-4 function, while Smad2 (also designated Madr2 or JV18-1) and Smad3 are involved in TGFβ and Activin-mediated growth modulation. Smad4 (also designated DPC4) has been shown to mediate all of the above activities through interaction with various Smad family members. Smad6 and Smad7 regulate the response to Activin/TGFβ signaling by interfering with TGFβ-mediated phosphorylation of other Smad proteins.

Immunogen: Synthetic phospho-peptide corresponding to residues surrounding Ser255 of Human Smad2 aa 229-272 / 467.

Positive control: HeLa cell lysate, HeLa treated with 200nM Calyculin A for 1 hour cell lysate, RAW264.7 cell lysate, mouse uterus tissue.

Subcellular location: Nucleus, Cytoplasm.

Database links: SwissProt: Q15796 Human | Q62432 Mouse | O70436 Rat

Recommended Dilutions:

WB	1:1,000
IHC-P	1:50-1:200
IP	Use at an assay dependent concentration.

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

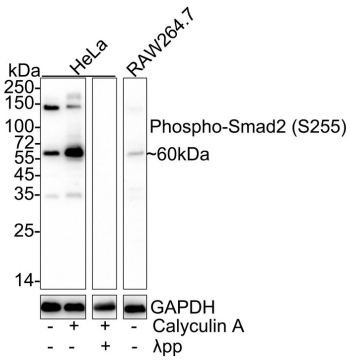

Fig1: Western blot analysis of Phospho-Smad2 (S255) on different lysates with Rabbit anti-Phospho-Smad2 (S255) antibody (ET1702-34) at 1/1,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HeLa treated with 200nM Calyculin A for 1 hour cell lysate
Lane 3: HeLa treated with 200nM Calyculin A for 1 hour cell lysate, then the membrane treated with λpp for 1 hour
Lane 4: RAW264.7 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 52 kDa
Observed band size: 60 kDa

Exposure time: 3 minutes; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1702-34) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

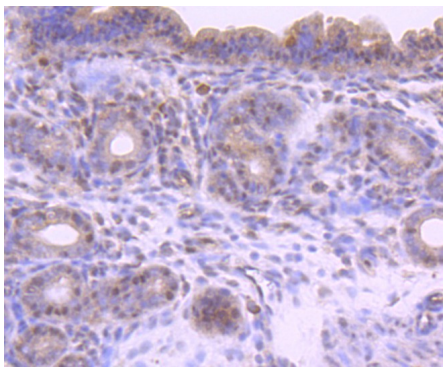

Fig2: Immunohistochemical analysis of paraffin-embedded mouse uterus tissue using anti-Phospho-Smad2 (S255) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-34, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Background References

1. Ungefroren H et al. Rac1b negatively regulates TGF- 1-induced cell motility in pancreatic ductal epithelial cells by suppressing Smad signalling. Oncotarget 5:277-90 (2014).
2. Harazono Y et al. miR-655 Is an EMT-suppressive MicroRNA targeting ZEB1 and TGFBR2. PLoS One 8:e62757 (2013).