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Phospho-Smad1/5-S463/465 Rabbit mAb

Catalog No.: AP1653 Recombinant

Basic Information

Observed MW
60 kDa

Calculated MW
15 kDa/52 kDa

Category
Primary antibody

Applications
WB,IF/ICC,ELISA

Cross-Reactivity
Human, Mouse, Rat

CloneNo number
ARC4094

Background

The protein encoded by this gene belongs to the SMAD, a family of proteins similar to the gene products of the Drosophila gene 'mothers against decapentaplegic' (Mad) and the C. elegans gene Sma. SMAD proteins are signal transducers and transcriptional modulators that mediate multiple signaling pathways. This protein mediates the signals of the bone morphogenetic proteins (BMPs), which are involved in a range of biological activities including cell growth, apoptosis, morphogenesis, development and immune responses. In response to BMP ligands, this protein can be phosphorylated and activated by the BMP receptor kinase. The phosphorylated form of this protein forms a complex with SMAD4, which is important for its function in the transcription regulation. This protein is a target for SMAD-specific E3 ubiquitin ligases, such as SMURF1 and SMURF2, and undergoes ubiquitination and proteasome-mediated degradation. Alternatively spliced transcript variants encoding the same protein have been observed.

Recommended Dilutions

WB	1:3000 - 1:15000
IF/ICC	1:1000 - 1:2000
ELISA	Recommended starting concentration is 1 μg/mL. Please optimize the concentration based on your specific assay requirements. For high-ratio antibody dilutions (≥1:10000)，a sequential dilution method is strongly recommended to ensure measurement accuracy.

Immunogen Information

Gene ID
4090/4086

Swiss Prot
Q99717/Q15797

Immunogen
This information is considered to be commercially sensitive.

Synonyms
DWFC; JV5-1; MADH5;BSP1; JV41; BSP-1; JV4-1; MADH1; MADR1

Product Information

Source
Rabbit

Isotype
IgG

Purification
Affinity purification

Storage
Store at -20℃. Avoid freeze / thaw cycles.
Buffer: PBS with 0.09% Sodium azide,0.05% BSA,50% glycerol,pH7.3.

Contact

400-999-6126

cn.market@abclonal.com.cn

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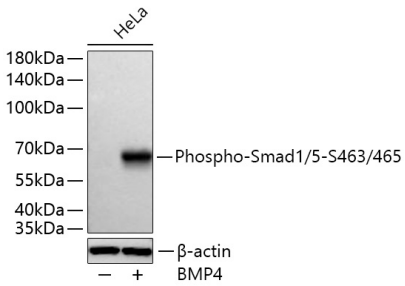
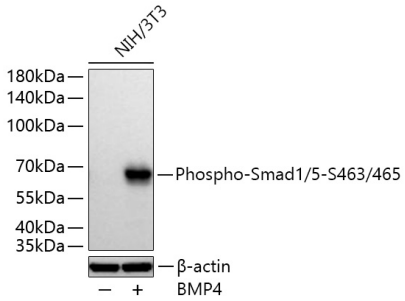
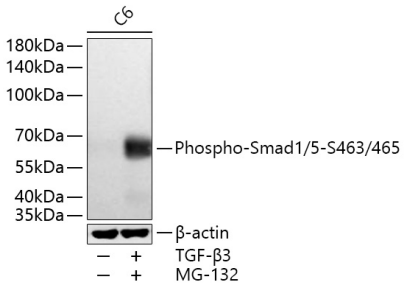
Validation Data



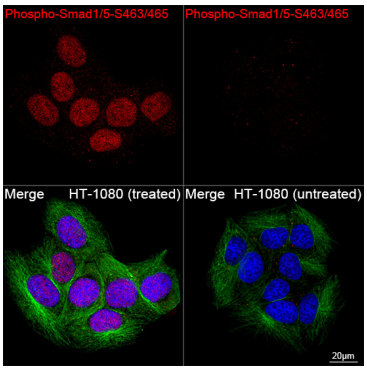
Western blot analysis of lysates from HeLa cells using Phospho-Smad1/5-S463/465 Rabbit mAb (AP1653) at 1:6000 dilution incubated overnight at 4℃. HeLa cells were treated with BMP4 (50 μg/mL) at 37℃ for 30 minutes after serum starvation overnight.
Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.
Lysates/proteins: 30 μg per lane.
Blocking buffer: 3% nonfat dry milk in TBST.
Detection: ECL Basic Kit (RM00020).
Exposure time: 10 s.



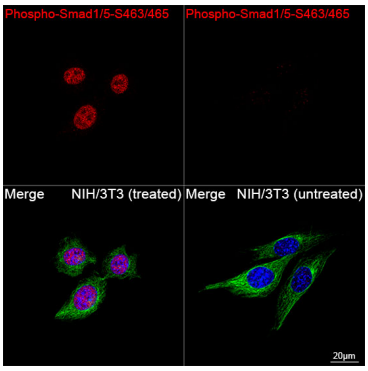
Western blot analysis of lysates from NIH/3T3 cells using Phospho-Smad1/5-S463/465 Rabbit mAb (AP1653) at 1:6000 dilution incubated overnight at 4℃. NIH/3T3 cells were treated with BMP4 (50 μg/mL) at 37℃ for 30 minutes after serum starvation overnight.
Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.
Lysates/proteins: 30 μg per lane.
Blocking buffer: 3% nonfat dry milk in TBST.
Detection: ECL Basic Kit (RM00020).
Exposure time: 10 s.



Western blot analysis of lysates from C6 cells using Phospho-Smad1/5-S463/465 Rabbit mAb (AP1653) at 1:6000 dilution incubated overnight at 4℃. C6 cells were treated with TGF-β3 (50 ng/mL) at 37℃ for 30 minutes and MG-132 (10 μM) at 37℃ for 30 minutes after serum starvation overnight..
Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.
Lysates/proteins: 30 μg per lane.
Blocking buffer: 3% nonfat dry milk in TBST.
Detection: ECL Basic Kit (RM00020).
Exposure time: 10 s.

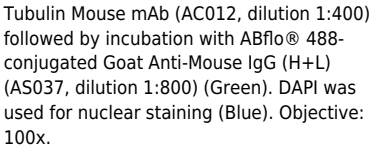


Confocal imaging of HT-1080 cells (treated with BMP4) and HT-1080 cells (untreated) using Phospho-Smad1/5-S463/465 Rabbit mAb (AP1653, dilution 1:2000) followed by a further incubation with Cy3-conjugated Goat anti-Rabbit IgG (H+L) (AS007, dilution 1:500) (Red). The cells were counterstained with α-

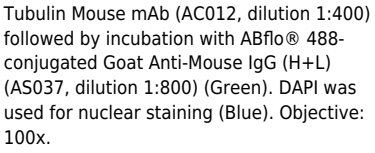


Confocal imaging of NIH/3T3 cells (treated with BMP4) and NIH/3T3 cells (untreated) using Phospho-Smad1/5-S463/465 Rabbit mAb (AP1653, dilution 1:2000) followed by a further incubation with Cy3-conjugated Goat anti-Rabbit IgG (H+L) (AS007, dilution 1:500) (Red). The cells were counterstained with α-

Validation Data



Tubulin Mouse mAb (AC012, dilution 1:400) followed by incubation with ABflo® 488-conjugated Goat Anti-Mouse IgG (H+L) (AS037, dilution 1:800) (Green). DAPI was used for nuclear staining (Blue). Objective: 100x.



Tubulin Mouse mAb (AC012, dilution 1:400) followed by incubation with ABflo® 488-conjugated Goat Anti-Mouse IgG (H+L) (AS037, dilution 1:800) (Green). DAPI was used for nuclear staining (Blue). Objective: 100x.