Smad2 [pT8] ABfinity™ Recombinant Rabbit Monoclonal Antibody - Purified

Catalog no. 700050

(See product label for lot information)

Clone/PAD: 11H10L30
Isotype: IgG
Gene ID: 4087
Protein Acc. no.: Q15796
Qty: 100 µg
Volume: 200 µl
Concentration: 0.5 mg/mL

Formulation
PBS + 0.09% sodium azide

Validation
Validated for use in WB

Reactivity
This antibody is specific for human Smad2 phosphorylated at pT8 and does not recognize non-phosphorylated Smad2 protein.

Immunogen
peptide

Immunogen sequence
ILPF[pT]PPVVKRL

Sequence Identity
Bovine, rat, drosophila, xenopus, mouse

Expected Reactivity
Based on sequence identity and similarity, reactivity to bovine, rat, drosophila, xenopus, and mouse is expected.

Storage
2-8°C for up to 1 month, -20°C for long term storage. Avoid repeated freezing and thawing.

Expiration Date
Expires one year from date of receipt when stored as instructed.

Background
SMAD2, also known as MADH2 or MAD2 regulates multiple cellular processes, such as cell proliferation, apoptosis, and differentiation (1). Smad2 interacts with the TGF-beta receptors (2, 3) through its interaction with the SMAD anchor for receptor activation (SARA) protein. The regulation of Smad2 phosphorylation on threonine 8 by ERK1 (4-7) and calmodulin is critical for Smad2-mediated signaling. Activated Smad2 associates with Smad4 and translocates as a complex into the nucleus, allowing its binding to DNA and transcription factors and thereby participating in the regulaton of gene expression. This translocation of Smad2 (as well as Smad3) into the nucleus is a central event in TGFβ signaling. Phosphorylation of threonine 8 in the calmodulin-binding region of the MH1 domain by extracellular signal-regulated kinase 1 (ERK1) enhances Smad2 transcriptional activity, which is negatively regulated by calmodulin

References

1. Cao, Z., et al. (2003) Levels of phospho-Smad2/3 are sensors of the interplay between effects of TGF-beta and retinoic acid on monocytic and granulocytic differentiation of HL-60 cells. Blood. 101(2):498-507.
2. Cordenonsi, M., et al. (2003) Links between tumor suppressors. p53 Is required for TGF-beta gene responses by cooperating with Smads. Cell. 113(3):301-314.
3. Hayashida, T., et al. (2003) Cross-talk between ERK MAP kinase and Smad signaling pathways enhances TGF-beta-dependent responses in human mesangial cells. FASEB J. 17(11):1576-1578.
4. Funaba, M., et al. (2002) Modulation of Smad2-mediated signaling by extracellular signal-regulated kinase. J. Biol. Chem. 277(44):41361-41368.
5. Blanchette, F., et al. (2001) Cross-talk between the p42/p44 MAP kinase and Smad pathways in transforming growth factor beta 1-induced furin gene transactivation. J. Biol. Chem. 276(36):33986-33994.
6. Brown, J.D., et al. (1999) MEKK-1, a component of the stress (stress-activated protein kinase/c-Jun N-terminal kinase) pathway, can selectively activate Smad2-mediated transcriptional activation in endothelial cells. J. Biol. Chem. 274(13):8797-8805.
7. Nakao, A., et al. (1997) Identification of Smad2, a human Mad-related protein in the transforming growth factor beta signaling pathway. J. Biol. Chem. 272(5):2896-2900.

Following applications had been tested during development. To make sure the consistency and reliability in the future lots, each lot is tested with antigen ELISA for specificity and potency. Each lot is also tested with SDS-PAGE, to ensure high purity.

Applications:

	Species	Test Material	Concentration
Western Blotting	human	HeLa + TGF-β	1-5 µg/ml

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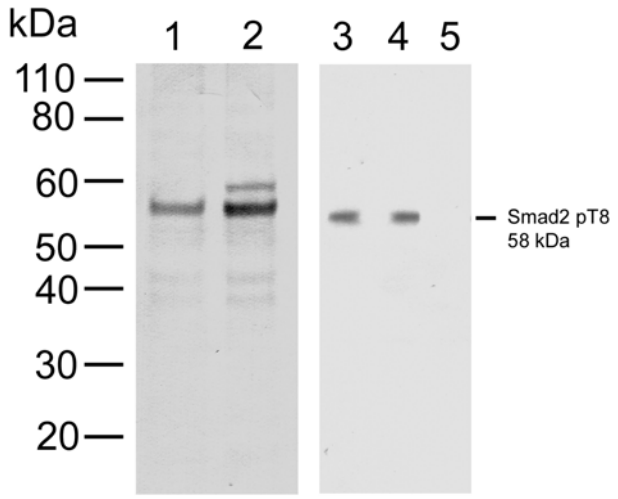

Western blot of HeLa cell lysate treated with TGF-β using rabbit anti-Smad2 pT8 (Cat. No. 700050).

Rabbit anti-Smad2 pT8 (1 µg/ml) was used to label phosphorylated Smad2 in TGF-β treated HeLa cell lysate. Phosphorylation of Smad2 at threonine 8 is increasing upon treatment of HeLa cells. Cells were serum starved over night (lane1) before addition of TGF-β (10 ng/ml for 30 min) (lane 2). Lane 3-5 represent competition experiments on lysate from HeLa cells treated with TGF-□as before. The antibody was used at 1 □g/ml no peptide (lane 3), antibody was preinicubated with the non-phospho peptide (lane 4) or preincubated with the phospho-peptide (lane 5) to show antibody specificity to the pT8 site. Each lane was loaded with 30 □g of lysate. The western was performed using the WesternBreeze® kit with NBT/BCIP as the substrate (Cat. No.WB7105).

Explanation of symbols

Symbol	Description	Symbol	Description
REF	Catalogue Number	LOT	Batch code
RUO	Research Use Only	IVD	*In vitro* diagnostic medical device
	Use by		Temperature limitation
	Manufacturer	EC REP	European Community authorised representative
[-]	Without, does not contain	[+]	With, contains
	Protect from light		Consult accompanying documents
	Directs the user to consult instructions for use (IFU), accompanying the product.		

For research use only. CAUTION: Not for human or animal therapeutic or diagnostic use.

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