Revision 1

Store at -20C

#8769

Phospho-Smad3 (Ser423/425) (D12E11) Rabbit mAb

Cell Signaling TECHNOLOGY®

Orders: 877-616-CELL (2355) orders@cellsignal.com

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Web: info@cellsignal.com cellsignal.com

3 Trask Lane | Danvers | Massachusetts | 01923 | USA

For Research Use Only. Not for Use in Diagnostic Procedures.

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
W, IP, IF-IC, FC-FP	H M R Mk	Endogenous	52	Rabbit IgG	#P84022	4088

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:50
Immunofluorescence (Immunocytochemistry)	1:250
Flow Cytometry (Fixed/Permeabilized)	1:200

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.

Specificity/Sensitivity

Phospho-Smad3 (Ser423/425) (D12E11) Rabbit mAb recognizes endogenous levels of Smad3 protein when phosphorylated at Ser422 only, at both Ser423 and Ser425, or at Ser422, Ser423, and Ser425. This antibody also weakly recognizes the equivalent phospho-residues of Smad2 protein (Ser464, Ser465, and Ser467).

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser422/423/425 of human Smad3 protein.

Background

Members of the SMAD family of signal transduction molecules are components of a critical intracellular pathway that transmit TGF-β signals from the cell surface into the nucleus. Three distinct classes of SMADs have been defined: the receptor-regulated SMADs (R-SMADs), which include SMAD1, 2, 3, 5, and 9; the common-mediator SMAD (co-SMAD), SMAD4; and the antagonistic or inhibitory SMADs (I-SMADs), SMAD6 and 7 (1-5). Activated type I receptors associate with specific R-SMADs and phosphorylate them on a conserved carboxy-terminal SSXS motif. The phosphorylated R-SMADs dissociate from the receptor and form a heteromeric complex with SMAD4, initiating translocation of the heteromeric SMAD complex to the nucleus. Once in the nucleus, SMADs recruit a variety of DNA binding proteins that function to regulate transcriptional activity (6-8).

Background References

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2. Attisano, L. and Wrana, J.L. (1998) *Curr Opin Cell Biol* 10, 188-94.
3. Derynck, R. et al. (1998) *Cell* 95, 737-40.
4. Massagué, J. (1998) *Annu Rev Biochem* 67, 753-91.
5. Whitman, M. (1998) *Genes Dev* 12, 2445-62.
6. Wrana, J.L. (2000) *Sci STKE* 2000, re1.
7. Attisano, L. and Wrana, J.L. (2002) *Science* 296, 1646-7.
8. Moustakas, A. et al. (2001) *J Cell Sci* 114, 4359-69.

Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

Western Blot Buffer

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

Applications Key

W: Western Blotting **IP:** Immunoprecipitation **IF-IC:** Immunofluorescence (Immunocytochemistry) **FC-FP:** Flow Cytometry (Fixed/Permeabilized)

Cross-Reactivity Key

H: Human **M:** Mouse **R:** Rat **Mk:** Monkey

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