# Smad3 siRNA (r): sc-77326



The Power to Question

#### **BACKGROUND**

Smad proteins, the mammalian homologs of the <code>Drosophila</code> mothers against dpp (Mad) have been implicated as downstream effectors of TGF $\beta$ /BMP signaling. Smad1 (also designated Madr1 or JV4-1), Smad5 and mammalian Smad8 (also designated Smad9 or MADH6) are effectors of BMP2 and BMP4 function while Smad2 (also designated Madr2 or JV18-1) and Smad3 are involved in TGF $\beta$  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/TGF $\beta$  signaling by interfering with TGF $\beta$ -mediated phosphorylation of other Smad family members.

#### **REFERENCES**

- 1. Liu, F., et al. 1996. A human Mad protein acting as a BMP-regulated transcriptional activator. Nature 381: 620-623.
- Eppert, K., et al. 1996. MADR2 maps to 18q21 and encodes a TGFβ-regulated MAD-related protein that is functionally mutated in colorectal carcinoma. Cell 86: 543-552.
- 3. Zhang, Y., et al. 1996. Receptor-associated Mad homologues synergize as effectors of the TGF $\beta$  response. Nature 383: 168-172.
- 4. Lagna, G., et al. 1996. Partnership between DPC4 and Smad proteins in  $TGF\beta$  signalling pathways. Nature 383: 832-836.

#### **CHROMOSOMAL LOCATION**

Genetic locus: Smad3 (rat) mapping to 8q24.

## **PRODUCT**

Smad3 siRNA (r) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10  $\mu M$  solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Smad3 shRNA Plasmid (r): sc-77326-SH and Smad3 shRNA (r) Lentiviral Particles: sc-77326-V as alternate gene silencing products.

For independent verification of Smad3 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-77326A, sc-77326B and sc-77326C.

# STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330  $\mu$ l of the RNAse-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNAse-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## **APPLICATIONS**

 $\mbox{Smad3}$  siRNA (r) is recommended for the inhibition of  $\mbox{Smad3}$  expression in rat cells.

#### **SUPPORT REAGENTS**

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

#### **GENE EXPRESSION MONITORING**

Smad2/3 (C-8): sc-133098 is recommended as a control antibody for monitoring of Smad3 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

#### **RT-PCR REAGENTS**

Semi-quantitative RT-PCR may be performed to monitor Smad3 gene expression knockdown using RT-PCR Primer: Smad3 (r)-PR: sc-77326-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

#### **SELECT PRODUCT CITATIONS**

- Watkins, S.J., et al. 2012. Angiotensin II-induced cardiomyocyte hypertrophy in vitro is TAK1-dependent and Smad2/3-independent. Hypertens. Res. 35: 393-398.
- Wang, S., et al. 2012. Up-regulation of BMP-2 antagonizes TGF-β1/ROCKenhanced cardiac fibrotic signalling through activation of Smurf1/Smad6 complex. J. Cell. Mol. Med. 16: 2301-2310.
- 3. Hedrick, E.D., et al. 2013. Differential PKA activation and AKAP association determines cell fate in cancer cells. J. Mol. Signal. 8: 10.
- 4. Wang, X., et al. 2015. NF $\kappa$ B inhibitor reverses temozolomide resistance in human glioma TR/U251 cells. Oncol. Lett. 9: 2586-2590.
- 5. Liu, X.H., et al. 2016. NADPH oxidase 4 contributes to connective tissue growth factor expression through Smad3-dependent signaling pathway. Free Radic. Biol. Med. 94: 174-184.
- Muscella, A., et al. 2019. TGF-β1 activates RSC96 Schwann cells migration and invasion through MMP-2 and MMP-9 activities. J. Neurochem. 153: 525-538.

## **RESEARCH USE**

For research use only, not for use in diagnostic procedures.

# **PROTOCOLS**

See our web site at www.scbt.com for detailed protocols and support products.