

TGIF siRNA (m): sc-36660

BACKGROUND

TGIF (5'-TG-3' interacting factor) was originally identified as a homeo-domain protein that binds to a retinoid X receptor (RXR) responsive element, thereby inhibiting the binding of RXR to this site and repressing RXR-dependent transcriptional activation. TGIF is a member of the TALE (three amino acid loop extension) family of homeodomain-containing proteins. TGIF also binds to Smad2, to repress Smad2-Smad4-mediated transcription. Smad2, after phosphorylation mediated by TGF β receptor, forms a complex with Smad4 and enters the nucleus to regulate transcription. The Smad2-Smad4 complex can interact with coactivators to form a transcriptional activation complex. Alternatively, the Smad2-Smad4 complex can interact with TGIF and HDACs to form a transcriptional repressor complex. Upon interaction with Smad2, TGIF is recruited to TGF β -responsive genes, where it acts to repress TGF β -induced transcription.

REFERENCES

1. Bertolino, E., et al. 1995. A novel homeobox protein which recognizes a TGT core and functionally interferes with a retinoid-responsive motif. *J. Biol. Chem.* 270: 31178-31188.
2. Baker, J.C., et al. 1996. A novel mesoderm inducer, Madr2, functions in the activin signal transduction pathway. *Genes Dev.* 10: 1880-1889.
3. Janknecht, R., et al. 1996. TGF β -stimulated cooperation of Smad proteins with the coactivators CBP/p300. *Genes Dev.* 12: 2114-2119.
4. Lana, G., et al. 1996. Partnership between DPC4 and Smad proteins in TGF β signalling pathways. *Nature* 383: 832-836.
5. Burglin, T.R., et al. 1997. Analysis of TALE superclass homeobox genes (MEIS, PBC, KNOX, Iroquois, TGIF) reveals a novel domain conserved between plants and animals. *Nucleic Acids Res.* 25: 4173-4180.
6. Pouppnot, C., et al. 1998. Physical and functional interaction of Smads and p300/CBP. *J. Biol. Chem.* 273: 22865-22868.
7. Wotton, D., et al. 1999. A Smad transcriptional corepressor. *Cell* 97: 29-39.
8. Gripp, K.W., et al. 2000. Mutations in TGIF cause holoprosencephaly and link NODAL signalling to human neural axis determination. *Nat. Genet.* 25: 205-208.

CHROMOSOMAL LOCATION

Genetic locus: Tgif1 (mouse) mapping to 17 E1.3.

PRODUCT

TGIF siRNA (m) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TGIF shRNA Plasmid (m): sc-36660-SH and TGIF shRNA (m) Lentiviral Particles: sc-36660-V as alternate gene silencing products.

For independent verification of TGIF (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-36660A, sc-36660B and sc-36660C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

TGIF siRNA (m) is recommended for the inhibition of TGIF expression in mouse 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 μ M in 66 μ l. 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

TGIF (H-1): sc-17800 is recommended as a control antibody for monitoring of TGIF 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 TGIF gene expression knockdown using RT-PCR Primer: TGIF (m)-PR: sc-36660-PR (20 μ l, 579 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Mistry, D.S., et al. 2011. Gonadotropin-releasing hormone pulse sensitivity of follicle-stimulating hormone- β gene is mediated by differential expression of positive regulatory activator protein 1 factors and corepressors SKIL and TGIF1. *Mol. Endocrinol.* 25: 1387-1403.

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.