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TdTIF1 siRNA (m): sc-154171

BACKGROUND

Terminal deoxynucleotidyltransferase (TdT) is a DNA polymerase which catalyzes the addition of deoxyribonucleotides onto the 3'-hydroxyl end of DNA primers without a DNA template. TdT activity can be positively or negatively regulated by association with certain TdT-interacting factors. TdTIF1 (Terminal deoxynucleotidyltransferase-interacting factor 1), also called DNTTIP1 or TdIF1, localizes to the nucleus and functions to repress TdT activity. *In vitro*, TdTIF1 binds to the Pol β -like region of TdT, thus masking its DNA-binding region and blocking its access to the DNA. Expressed throughout the body with highest expression in the thymus, TdTIF1 contains N-terminal AT-hook motifs through which it binds to TdT. In addition to inhibiting TdT-DNA interaction, TdTIF1 can bind to TReP-132, a transcriptional co-activator of steroidogenic factor 1 (SF-1). When bound to SF-1, TdTIF1 enhances gene expression in steroid-producing cells.

REFERENCES

1. Yamashita, N., et al. 2001. Terminal deoxynucleotidyltransferase directly interacts with a novel nuclear protein that is homologous to p65. *Genes Cells* 6: 641-652.
2. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 611388. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Fujita, K., et al. 2003. Terminal deoxynucleotidyltransferase forms a ternary complex with a novel chromatin remodeling protein with 82 kDa and core histone. *Genes Cells* 8: 559-571.
4. Fujisaki, S., et al. 2005. Direct binding of TReP-132 with TdT results in reduction of TdT activity. *Genes Cells* 11: 47-57.
5. Kubota, T., et al. 2007. Identification of functional domains in TdIF1 and its inhibitory mechanism for TdT activity. *Genes Cells* 12: 941-959.

CHROMOSOMAL LOCATION

Genetic locus: Dnttip1 (mouse) mapping to 2 H3.

PRODUCT

TdTIF1 siRNA (m) is a pool of 2 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 TdTIF1 shRNA Plasmid (m): sc-154171-SH and TdTIF1 shRNA (m) Lentiviral Particles: sc-154171-V as alternate gene silencing products.

For independent verification of TdTIF1 (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-154171A and sc-154171B.

PROTOCOLS

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

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

TdTIF1 siRNA (m) is recommended for the inhibition of TdTIF1 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

TdTIF1 (G-2): sc-166296 is recommended as a control antibody for monitoring of TdTIF1 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).

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker[™] compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker[™] Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz[™] Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor TdTIF1 gene expression knockdown using RT-PCR Primer: TdTIF1 (m)-PR: sc-154171-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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