SANTA CRUZ BIOTECHNOLOGY, INC.

TTF siRNA (m): sc-38603

BACKGROUND

The transcription termination factor TTF (RNA polymerase I, TTF1, TTF-I) exerts two functions in ribosomal gene (rDNA) transcription: facilitating initiation and mediating termination of transcription. Sequence-specific termination of DNA replication within mammalian ribosomal RNA genes is catalyzed by a DNA-protein complex that includes TTF. Mammalian ribosomal genes are flanked at their 5' and 3' ends by terminator sequences which are recognized by the transcription termination factor TTF. In HeLa cells, TTF protein co-localizes with the active transcription machinery in the nucleolus and also with the inactive machinery present in certain mitotic nucleolar organizer regions (NORs) when rDNA transcription is repressed.

REFERENCES

1. Kuhn, A., et al. 1990. Specific interaction of the murine transcription termination factor TTF-I with class-I RNA polymerases. Nature 344: 559-562.
2. Evers, R., et al. 1995. Molecular coevolution of mammalian ribosomal gene terminator sequences and the transcription termination factor TTF-I. Proc. Natl. Acad. Sci. USA 92: 5827-5831.
3. Sander, E.E., et al. 1996. The amino-terminal domain of the transcription termination factor TTF-I causes protein oligomerization and inhibition of DNA binding. Nucleic Acids Res. 24: 3677-3684.
4. Langst, G., et al. 1997. RNA polymerase I transcription on nucleosomal templates: the transcription termination factor TTF-I induces chromatin remodeling and relieves transcriptional repression. EMBO J. 16: 760-768.
5. Sander, E.E., et al. 1997. Oligomerization of the transcription termination factor TTF-I: implications for the structural organization of ribosomal transcription units. Nucleic Acids Res. 25: 1142-1147.
6. Gerber, J.K., et al. 1997. Termination of mammalian rDNA replication: polar arrest of replication fork movement by transcription termination factor TTF-I. Cell 90: 559-567.
7. Sirri, V., et al. 1999. The mitotically phosphorylated form of the transcription termination factor TTF-1 is associated with the repressed rDNA transcription machinery. J. Cell Sci. 112: 3259-3268.

CHROMOSOMAL LOCATION

Genetic locus: Ttf1 (mouse) mapping to 2 A3.

PRODUCT

TTF 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TTF shRNA Plasmid (m): sc-38603-SH and TTF shRNA (m) Lentiviral Particles: sc-38603-V as alternate gene silencing products.

For independent verification of TTF (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-38603A, sc-38603B and sc-38603C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

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

TTF (A-5): sc-398968 is recommended as a control antibody for monitoring of TTF 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 reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor TTF gene expression knockdown using RT-PCR Primer: TTF (m)-PR: sc-38603-PR (20 µl). 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.

PROTOCOLS

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