SANTA CRUZ BIOTECHNOLOGY, INC.

TFII-I siRNA (m): sc-36644

BACKGROUND

Initiation of transcription of eukaryotic genes requires the association of large multimeric protein complexes that involve RNA polymerase II and a variety of basal transcription factors, including members of the TFII protein family. TFII proteins complex with Pol II and initiate transcription by binding to the core promoter elements, such as TATA box sequences, that are located upstream of the transcription start codon. A member of the TFII family, TFII-I is regulated by tyrosine phophorylation, and it is involved in the initiation of transcription of TATA-less promoters and in cell type specific transcription. TFII-I directly associates with several promoters elements, including TATA box, pyrimidine-rich initiator (Inr) and E-box elements. TFII-I is also implicated in activating transcription of the transcription factor c-Fos, a downstream effector protein in the MAP kinase-signaling pathway. TFII-I binds to several initiation sites within the c-Fos promoter, and it is phosphorylated in response to Map kinase activation, which then enhances TFII-I induced expression of c-Fos.

REFERENCES

1. Conaway, R.C., et al. 1989. An RNA polymerase II transcription factor has an associated DNA-dependent ATPase (dATPase) activity strongly stimulated by the TATA region of promoters. Proc. Natl. Acad. Sci. USA 86: 7356-7360.
2. Roy, A.L., et al. 1993. An alternative pathway for transcription initiation involving TFII-I. Nature 365: 355-359.
3. Holstege, F.C., et al. 1998. Dissecting the regulatory circuitry of a eukaryotic genome. Cell 95: 717-728.
4. Majello, B., et al. 1998. Recruitment of human TBP selectively activates RNA polymerase II TATA-dependent promoters. J. Biol. Chem. 273: 16509-16516.
5. Novina, C.D., et al. 1998. Regulation of TFII-I activity by phosphorylation. J. Biol. Chem. 273: 33443-33448.
6. Kim, D.W., et al. 1998. TFII-I enhances activation of the c-Fos promoter through interactions with upstream elements. Mol. Cell. Biol. 18: 3310-3320.
7. Cheriyath, V., et al. 1998. TFII-I regulates Vβ promoter activity through an initiator element. Mol. Cell. Biol. 18: 4444-4454.

CHROMOSOMAL LOCATION

Genetic locus: Gtf2i (mouse) mapping to 5 G2.

PRODUCT

TFII-I 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 TFII-I shRNA Plasmid (m): sc-36644-SH and TFII-I shRNA (m) Lentiviral Particles: sc-36644-V as alternate gene silencing products.

For independent verification of TFII-I (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-36644A, sc-36644B and sc-36644C.

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

TFII-I siRNA (m) is recommended for the inhibition of TFII-I 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

TFII-I (B-7): sc-46670 is recommended as a control antibody for monitoring of TFII-I 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 TFII-I gene expression knockdown using RT-PCR Primer: TFII-I (m)-PR: sc-36644-PR (20 µl, 457 bp). 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.