SANTA CRUZ BIOTECHNOLOGY, INC.

TFIIF RAP 30 siRNA (m): sc-38522

BACKGROUND

In eukaryotic systems, initiation of transcription from protein-coding genes is a complex process requiring RNA polymerase II and broad families of auxiliary transcription factors. Such factors can be divided into two major functional classes: the basal factors that are required for transcription of all Pol II genes, including TFIIA, TFIIB, TFIID, TFIIE, TFIIF and TFIIH; and sequence-specific factors that regulate gene expression. The basal transcription factors and Pol II form a specific multiprotein complex near the transcription start site by interacting with core promotor elements such as the TATA box generally located 25-30 base pairs upstream of the transcription start site. TFIIF, a heteromer composed of a small (RAP 30) and a large (RAP 74) subunit, is required for RNA polymerase II to assemble into a preinitiation complex formed by promotor DNA and the general factors TFIID, IIA and IIB. In addition, TFIIF stimulates transcription elongation by RNA polymerase II.

REFERENCES

1. Sopta, M., et al. 1989. Structure and associated DNA-helicase activity of a general transcription initiation factor that binds to RNA polymerase II. Nature 341: 410-414.
2. Maldonado, E., et al. 1990. Factors involved in specific transcription by mammalian RNA polymerase II: role of transcription factors IIA, IID, and IIB during formation of a transcription-competent complex. Mol. Cell. Biol. 10: 6335-6347.
3. Peterson, M.G., et al. 1990. Functional domains and upstream activation properties of cloned human TATA binding protein. Science 248: 1625-1630.
4. Peterson, M.G., et al. 1991. Structure and functional properties of human general transcription factor IIE. Nature 354: 369-373.
5. Lee, D.K., et al. 1992. TFIIA induces conformational changes in TFIID via interactions with the basic repeat. Mol. Cell. Biol. 12: 5189-5196.
6. Aso, T., et al. 1992. Characterization of cDNA for the large subunit of the transcription initiation factor TFIIF. Nature 355: 461-467.
7. Yonaha, M., et al. 1993. Domain structure of a human general transcription initiation factor, TFIIF. Nucleic Acids Res. 21: 273-279.

CHROMOSOMAL LOCATION

Genetic locus: Gtf2f2 (mouse) mapping to 14 D3.

PRODUCT

TFIIF RAP 30 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 TFIIF RAP 30 shRNA Plasmid (m): sc-38522-SH and TFIIF RAP 30 shRNA (m) Lentiviral Particles: sc-38522-V as alternate gene silencing products.

For independent verification of TFIIF RAP 30 (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-38522A, sc-38522B and sc-38522C.

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

TFIIF RAP 30 siRNA (m) is recommended for the inhibition of TFIIF RAP 30 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

TFIIF RAP 30 (G-3): sc-374304 is recommended as a control antibody for monitoring of TFIIF RAP 30 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 TFIIF RAP 30 gene expression knockdown using RT-PCR Primer: TFIIF RAP 30 (m)-PR: sc-38522-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.