SANTA CRUZ BIOTECHNOLOGY, INC.

TFIIA-γ siRNA (h): sc-36645

BACKGROUND

Initiation of transcription from protein-coding genes in eukaryotes is a complex process that requires RNA polymerase II, as well as families of basal transcription factors. Binding of the factor TFIID (TBP) to the TATA box is believed to be the first step in the formation of a multiprotein complex containing several additional factors, including TFIIA, TFIIB, TFIIE, TFIIF and TFIIH. Recognition of the TATA binding element by TBP, one of the first steps in transcription initiation, may be regulated by TFIIA. TFIIA consists of three subunits designated TFIIA-α, TFIIA-β and TFIIA-γ, and it interacts with both TBP and a TAF (TBP-associated factor). It has been demonstrated that the basic region of TBP is essential for TFIIA-dependent function of TBP.

REFERENCES

1. Nakajima, N., et al. 1988. Factors involved in specific transcription by mammalian RNA polymerase II: purification, genetic specificity, and TATA box-promoter interactions of TFIID. Mol. Cell. Biol. 8: 4028-4040.
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3. 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.
4. 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.
5. Gerard, M., et al. 1991. Purification and interaction properties of the human polymerase B (II) general transcription factor BTF2. J. Biol. Chem. 266: 20940-20945.
6. Flores, O., et al. 1992. Factors involved in specific transcription by mammalian RNA polymerase II. J. Biol. Chem. 267: 2786-2793.
7. Ozer, J., et al. 1994. Molecular cloning of the small (γ) subunit of human TFIIA reveals functions critical for activated transcription. Genes Dev. 8: 2324-2335.

CHROMOSOMAL LOCATION

Genetic locus: GTF2A2 (human) mapping to 15q22.2.

PRODUCT

TFIIA-γ siRNA (h) 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 TFIIA-γ shRNA Plasmid (h): sc-36645-SH and TFIIA-γ shRNA (h) Lentiviral Particles: sc-36645-V as alternate gene silencing products.

For independent verification of TFIIA-γ (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-36645A, sc-36645B and sc-36645C.

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

TFIIA-γ siRNA (h) is recommended for the inhibition of TFIIA-γ expression in human 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

TFIIA-γ (D-6): sc-374483 is recommended as a control antibody for monitoring of TFIIA-γ 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 TFIIA-γ gene expression knockdown using RT-PCR Primer: TFIIA-γ (h)-PR: sc-36645-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.