

TFIIE- α siRNA (h): sc-36651

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 promoter elements such as the TATA box generally located 25-30 base pairs upstream of the transcription start site. Human TFIIE consists of two subunits, α and β . The structure of TFIIE appears to be a heterotrimer ($\alpha 2\beta 2$); both subunits are required for optimal basal-level transcription.

REFERENCES

1. 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.
2. Peterson, M.G., et al. 1990. Functional domains and upstream activation properties of cloned human TATA binding protein. *Science* 248: 1625-1630.

CHROMOSOMAL LOCATION

Genetic locus: GTF2E1 (human) mapping to 3q13.33.

PRODUCT

TFIIE- α 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TFIIE- α shRNA Plasmid (h): sc-36651-SH and TFIIE- α shRNA (h) Lentiviral Particles: sc-36651-V as alternate gene silencing products.

For independent verification of TFIIE- α (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-36651A, sc-36651B and sc-36651C.

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

TFIIE- α siRNA (h) is recommended for the inhibition of TFIIE- α expression in human cells.

RESEARCH USE

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

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

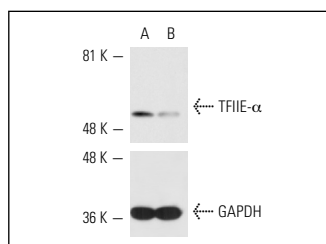
TFIIE- α (E-2): sc-133064 is recommended as a control antibody for monitoring of TFIIE- α gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 TFIIE- α gene expression knockdown using RT-PCR Primer: TFIIE- α (h)-PR: sc-36651-PR (20 μ l). Annealing temperature for the primers should be 55-60 $^{\circ}$ C and the extension temperature should be 68-72 $^{\circ}$ C.

DATA



TFIIE- α siRNA (h): sc-36651. Western blot analysis of TFIIE- α expression in non-transfected control (A) and TFIIE- α siRNA transfected (B) HeLa cells. Blot probed with TFIIE- α (C-17): sc-237. GAPDH (FL-335): sc-25778 used as specificity and loading control.

PROTOCOLS

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