SANTA CRUZ BIOTECHNOLOGY, INC.

HIV-1 Tat-SF1 siRNA (h): sc-62468

BACKGROUND

HIV-1 Tat-SF1 (HIV-1 Tat-specific factor 1) is a phosphoprotein that plays a role in the process of transcriptional elongation. It is ubiquitously expressed and localizes to the nucleus. HIV-1 Tat-SF1 interacts with Tat, P-TEFb, TFIIF RAP 30, CA150, Spt5, Pol II and U snRNPs. It is structurally similar to CUS2 in yeast. HIV-1 Tat-SF1 contains an acidic C-terminal motif and two RNA recognition motif (RRM) domains that mediate its interaction with U snRNPs. HIV-1 Tat-SF1 forms a complex with U snRNP, thereby coupling transcription and splicing. HIV-1 Tat-SF1 expression is upregulated by the HIV-1 proteins Nef and gp120. It acts as a cofactor for the Tat-enhanced transcription of HIV-1 and is required, along with SPT5, for the activation of Tat. Overexpression of Tat-SF1 and SPT5 stimulates the transcriptional activity of Tat.

REFERENCES

1. Li, X.Y. and Green, M.R. 1998. The HIV-1 Tat cellular coactivator Tat-SF1 is a general transcription elongation factor. Genes Dev. 12: 2992-2996.
2. Parada, C.A. and Roeder, R.G. 1999. A novel RNA polymerase II-containing complex potentiates Tat-enhanced HIV-1 transcription. EMBO J. 18: 3688-3701.
3. Kim, J.B., et al. 1999. Tat-SF1 protein associates with RAP 30 and human SPT5 proteins. Mol. Cell. Biol. 19: 5960-5968.
4. Fong, Y.W. and Zhou, Q. 2000. Relief of two built-in autoinhibitory mechanisms in P-TEFb is required for assembly of a multicomponent transcription elongation complex at the human immunodeficiency virus type 1 promoter. Mol. Cell. Biol. 20: 5897-5907.
5. Simmons, A., et al. 2001. Nef triggers a transcriptional program in T cells imitating single-signal T cell activation and inducing HIV virulence mediators. Immunity 14: 763-777.
6. Fong, Y.W. and Zhou, Q. 2002. Stimulatory effect of splicing factors on transcriptional elongation. Nature 414: 929-933.
7. Sudbrak, R., et al. 2003. Comparative and evolutionary analysis of the rhesus macaque extended MHC class II region. Immunogenetics 54: 699-704.
8. Kameoka, S., et al. 2004. p54^{nrb} associates with the 5' splice site within large transcription/splicing complexes. EMBO J. 23: 1782-1791.

CHROMOSOMAL LOCATION

Genetic locus: HTATSF1 (human) mapping to Xq26.3.

PRODUCT

HIV-1 Tat-SF1 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 HIV-1 Tat-SF1 shRNA Plasmid (h): sc-62468-SH and HIV-1 Tat-SF1 shRNA (h) Lentiviral Particles: sc-62468-V as alternate gene silencing products.

For independent verification of HIV-1 Tat-SF1 (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-62468A, sc-62468B and sc-62468C.

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

HIV-1 Tat-SF1 siRNA (h) is recommended for the inhibition of HIV-1 Tat-SF1 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

HIV-1 Tat-SF1 (C-4): sc-514351 is recommended as a control antibody for monitoring of HIV-1 Tat-SF1 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 HIV-1 Tat-SF1 gene expression knockdown using RT-PCR Primer: HIV-1 Tat-SF1 (h)-PR: sc-62468-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.