SANTA CRUZ BIOTECHNOLOGY, INC.

Positive cofactor 4 siRNA (h): sc-38583

BACKGROUND

In eukaryotic cells, transcription is regulated in part by high molecular weight co-activating complexes that mediate signals between transcriptional activators and RNA polymerase. RNA polymerase II (RNAPII) holoenzyme contains numerous proteins that largely consist of RNA processing factors, RNA helicase, general transcription factors and SRB co-activating complexes. RNAPII mediated basal- and gene-specific transcriptional activation requires the association of various cofactors that includes PC4 (human Positive cofactor 4). Positive cofactor 4 interacts with the activation domain of transcription factor IIA (TFIIA) and TATA-binding protein (TBP)-associated factors (TAFs) to directly bind to double stranded DNA. Positive cofactor 4 induces both activation and repression of RNAPII basal transcription, depending on the presence or absence of these transcription factors and holoenzyme components. Additionally, Positive cofactor 4 is phosphorylated by TFIID and TFIIH, which releases Positive cofactor 4 from the DNA promoter region and thereby inhibits the assembly of Positive cofactor 4 into the transcriptional promoting complex and blocks transcription.

REFERENCES

1. Ge, H. and Roeder, R.G. 1994. Purification, cloning, and characterization of a human coactivator, PC4, that mediates transcriptional activation of class II genes. Cell 78: 513-523.
2. Kaiser, K., et al. 1995. The co-activator p15 (PC4) initiates transcriptional activation during TFIIA-TFIID-promoter complex formation. EMBO J. 14: 3520-3527.

CHROMOSOMAL LOCATION

Genetic locus: SUB1 (human) mapping to 5p13.3.

PRODUCT

Positive cofactor 4 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 Positive cofactor 4 shRNA Plasmid (h): sc-38583-SH and Positive cofactor 4 shRNA (h) Lentiviral Particles: sc-38583-V as alternate gene silencing products.

For independent verification of Positive cofactor 4 (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-38583A, sc-38583B and sc-38583C.

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

Positive cofactor 4 siRNA (h) is recommended for the inhibition of Positive cofactor 4 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

Positive cofactor 4 (H-12): sc-166280 is recommended as a control antibody for monitoring of Positive cofactor 4 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 Positive cofactor 4 gene expression knockdown using RT-PCR Primer: Positive cofactor 4 (h)-PR: sc-38583-PR (20 µl, 484 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Hu, X., et al. 2017. Down regulation of human positive coactivator 4 suppress tumorigenesis and lung metastasis of osteosarcoma. Oncotarget 8: 53210-53225.

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.