

PCAF siRNA (m): sc-36199

BACKGROUND

In the intact cell, DNA closely associates with histones and other nuclear proteins to form chromatin. The remodeling of chromatin is believed to be a critical component of transcriptional regulation and a major source of this remodeling is brought about by the acetylation of nucleosomal histones. Acetylation of lysine residues in the amino terminal tail domain of histone results in an allosteric change in the nucleosomal conformation and an increased accessibility to transcription factors by DNA. Conversely, the deacetylation of histones is associated with transcriptional silencing. Several mammalian proteins have been identified as nuclear histone acetylases, including GCN5, PCAF (for p300/CBP-associated factor), p300/CBP and the TFIID subunit TAF II p250. Mammalian HDAC1 (also designated HD1) and HDAC2 (also designated mammalian RPD3), both of which are related to the yeast transcriptional regulator Rpd3p, have been identified as histone deacetylases.

REFERENCES

1. Lee, D.Y., et al. 1993. A positive role for histone acetylation in transcription factor access to nucleosomal DNA. *Cell* 72: 73-82.
2. Braunstein, M., et al. 1993. Transcriptional silencing in yeast is associated with reduced nucleosome acetylation. *Genes Dev.* 7: 592-604.
3. Bauer, W.R., et al. 1994. Nucleosome structural changes due to acetylation. *J. Mol. Biol.* 236: 685-690.
4. Brownell, J.E., et al. 1996. *Tetrahymena* histone acetyltransferase A: a homolog to yeast Gcn5p linking histone acetylation to gene activation. *Cell* 84: 843-851.

CHROMOSOMAL LOCATION

Genetic locus: Kat2b (mouse) mapping to 17 C.

PRODUCT

PCAF 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PCAF shRNA Plasmid (m): sc-36199-SH and PCAF shRNA (m) Lentiviral Particles: sc-36199-V as alternate gene silencing products.

For independent verification of PCAF (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-36199A, sc-36199B and sc-36199C.

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

PCAF siRNA (m) is recommended for the inhibition of PCAF 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 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

PCAF (E-8): sc-13124 is recommended as a control antibody for monitoring of PCAF 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 Marker[™] 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 PCAF gene expression knockdown using RT-PCR Primer: PCAF (m)-PR: sc-36199-PR (20 μ l, 500 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Sharma, M., et al. 2007. Regulation of transcript elongation through cooperative and ordered recruitment of cofactors. *J. Biol. Chem.* 282: 20887-20896.
2. Jang, S.M., et al. 2015. KAT5-mediated SOX4 acetylation orchestrates chromatin remodeling during myoblast differentiation. *Cell Death Dis.* 6: e1857.

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.