

RbAp48 siRNA (m): sc-37963

BACKGROUND

In the intact cell, DNA is closely associated 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 of DNA to transcription factors. 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), HDAC2 (also designated RPD3) and HDAC3, all of which are related to the yeast transcriptional regulator Rpd3p, have been identified as histone deacetylases. The retinoblastoma binding proteins RbAp46 and RbAp48 have been identified as histone binding proteins, and they are components of the histone deacetylase complex.

REFERENCES

1. Braunstein, M., et al. 1993. Transcriptional silencing in yeast is associated with reduced nucleosome acetylation. *Genes Dev.* 7: 592-604.
2. Bauer, W.R., et al. 1994. Nucleosome structural changes due to acetylation. *J. Mol. Biol.* 236: 685-690.
3. Qian, Y.W., et al. 1995. Dual retinoblastoma-binding proteins with properties related to a negative regulator of Ras in yeast. *J. Biol. Chem.* 270: 25507-25513.
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.
5. Yang, X.J., et al. 1996. A p300/CBP-associated factor that competes with the adenoviral oncoprotein E1A. *Nature* 382: 319-324.
6. Yang, W.M., et al. 1996. Transcriptional repression by YY1 is mediated by interaction with a mammalian homolog of the yeast global regulator RPD3. *Proc. Natl. Acad. Sci. USA* 93: 12845-12850.

CHROMOSOMAL LOCATION

Genetic locus: Rbbp4 (mouse) mapping to 4 D2.2.

PRODUCT

RbAp48 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 RbAp48 shRNA Plasmid (m): sc-37963-SH and RbAp48 shRNA (m) Lentiviral Particles: sc-37963-V as alternate gene silencing products.

For independent verification of RbAp48 (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-37963A, sc-37963B and sc-37963C.

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

RbAp48 siRNA (m) is recommended for the inhibition of RbAp48 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

RbAp46/p48 (G-8): sc-373873 is recommended as a control antibody for monitoring of RbAp48 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 RbAp48 gene expression knockdown using RT-PCR Primer: RbAp48 (m)-PR: sc-37963-PR (20 μ l). 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.