SANTA CRUZ BIOTECHNOLOGY, INC.

CBP siRNA (m): sc-29243

BACKGROUND

Cyclic AMP-regulated gene expression frequently involves a DNA element designated the cAMP-regulated enhancer (CRE). Many transcription factors, including the protein CREB, which is activated as a result of phosphorylation by protein kinase A, bind to this element. It has been shown that protein kinase A-mediated CREB phosphorylation results in its binding to a nuclear protein designated CBP (for CREB-binding protein). These findings suggest that CBP has many of the properties expected of a CREB co-activator. Another high molecular weight transcriptional adapter protein, designated p300, is characterized by three cysteine- and histidine-rich regions, of which the most carboxy terminal region specifically binds the adenovirus E1A protein. p300 molecules lacking an intact E1A binding site bypass E1A repression even in the presence of high concentrations of E1A. Sequence analysis of CBP and p300 has revealed substantial homology, arguing that these proteins are members of a conserved family of co-activators.

REFERENCES

1. Chivra, J.C., et al. 1993. Phosphorylated CREB binds specifically to the nuclear protein CBP. Nature 365: 855-859.
2. Kwok, R.P.S., et al. 1993. Nuclear protein CBP is a coactivator for the transcription factor CREB. Nature 370: 223-229.
3. Eckner, R., et al. 1994. Molecular cloning and functional analysis of the adenovirus E1A-associated 330-kDa protein (p300) reveals a protein with properties of a transcriptional adaptor. Genes Dev. 8: 869-884.
4. Arany, Z., et al. 1994. E1A-associated p300 and CREB-associated CBP belong to a conserved family of coactivators. Cell 77: 799-800.

CHROMOSOMAL LOCATION

Genetic locus: Crebbp (mouse) mapping to 16 A1.

PRODUCT

CBP siRNA (m) is a pool of 4 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 CBP shRNA Plasmid (m): sc-29243-SH and CBP shRNA (m) Lentiviral Particles: sc-29243-V as alternate gene silencing products.

For independent verification of CBP (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-29243A, sc-29243B, sc-29243C and sc-29243D.

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

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

CBP (C-1): sc-7300 is recommended as a control antibody for monitoring of CBP 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 CBP gene expression knockdown using RT-PCR Primer: CBP (m)-PR: sc-29243-PR (20 µl, 591 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Meissner, J.D., et al. 2007. The p38α/β mitogen-activated protein kinases mediate recruitment of CREB-binding protein to preserve fast Myosin heavy chain IId/x gene activity in myotubes. J. Biol. Chem. 282: 7265-7275.
2. Lee, Y., et al. 2010. Coactivation of the CLOCK-BMAL1 complex by CBP mediates resetting of the circadian clock. J. Cell Sci. 123: 3547-3557.
3. Riascos-Bernal, D.F., et al. 2016. β-catenin C-terminal signals suppress p53 and are essential for artery formation. Nat. Commun. 7: 12389.

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.