SANTA CRUZ BIOTECHNOLOGY, INC.

CIR siRNA (h): sc-38213

BACKGROUND

Recombination signal binding protein Jκ (RBP-Jκ), also designated KBF2 or CBF1, is the mammalian homolog of the *Drosophila* suppressor of hairless (Su(H)), a protein involved in the development of the peripheral nervous system. RBP-Jκ is ubiquitously expressed in mammalian tissues and is involved in the regulation of gene expression. RBP-Jκ has been shown to directly interact with the intracellular domain of the cell surface receptor Notch1. Proteolytically cleaved Notch1 translocates to the nucleus, where it binds to DNA-bound RBP-Jκ and activates transcription of target genes. CIR (for CBF1 interacting corepressor) serves as a linker between RBP-Jκ and the histone deacetylase complex by binding to SAP30 and to histone deacetylase. CIR binding to RBP-Jκ results in transcriptional repression of Notch 1 target genes.

REFERENCES

1. Amakawa, R., et al. 1993. Human Jκ recombination signal binding protein gene (IGKJRB): comparison with its mouse homologue. Genomics 17: 306-315.
2. Oka, C., et al. 1995. Disruption of the mouse RBP-Jκ gene results in early embryonic death. Development 121: 3291-3301.
3. Waltzer, L., et al. 1995. RBP-Jκ repression activity is mediated by a co-repressor and antagonized by the Epstein-Barr virus transcription factor EBNA2. Nucleic Acids Res. 23: 4939-4945.
4. Tamura, K., et al. 1995. Physical interaction between a novel domain of the receptor Notch and the transcription factor RBP-Jκ/Su(H). Curr. Biol. 5: 1416-1423.
5. Hsieh, J.J., et al. 1996. Truncated mammalian Notch1 activates CBF1/RBP Jκ-repressed genes by a mechanism resembling that of Epstein-Barr virus EBNA2. Mol. Cell. Biol. 16: 952-959.
6. Hsieh, J.J., et al. 1999. CIR, a corepressor linking DNA binding factor CBF1 to the histone deacetylase complex. Proc. Natl. Acad. Sci. USA 96: 23-28.

CHROMOSOMAL LOCATION

Genetic locus: CIR1 (human) mapping to 2q31.1.

PRODUCT

CIR 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 CIR shRNA Plasmid (h): sc-38213-SH and CIR shRNA (h) Lentiviral Particles: sc-38213-V as alternate gene silencing products.

For independent verification of CIR (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-38213A, sc-38213B and sc-38213C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

CIR siRNA (h) is recommended for the inhibition of CIR 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

CIR (H-1): sc-514120 is recommended as a control antibody for monitoring of CIR 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 CIR gene expression knockdown using RT-PCR Primer: CIR (h)-PR: sc-38213-PR (20 µl, 515 bp). 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.