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CGGBP1 siRNA (m): sc-142304

BACKGROUND

Fragile X syndrome is the most frequent form of inherited mental retardation and is the result of transcriptional silencing of the FMR1 (fragile X mental retardation) gene on the X chromosome. The FMR1 gene contains a distinct CpG dinucleotide repeat located in the 5'-untranslated region of the gene which, in fragile X syndrome, is substantially amplified and subject to extensive methylation and enhanced transcriptional silencing. CGGBP1 (CGG triplet repeat binding protein 1), also known as CGGBP or p20-CGGBP, is a 167 amino acid nuclear protein that influences FMR1 expression. Highly expressed in thymus, placenta, lymph nodes, cerebral cortex and cerebellum, CGGBP1 binds to the 5' (CGG)_n-3' repeat in the promotor of the FMR1 gene and positively regulates expression of the FMR1 protein. Binding of CGGBP1 to the FMR1 promoter is inhibited by cytosine-specific DNA methylation of the protein binding motif, suggesting that CGGBP1 activity is silenced in FMR1-affected individuals.

REFERENCES

1. Verkerk, A.J., et al. 1991. Identification of a gene (FMR-1) containing a CGG repeat coincident with a breakpoint cluster region exhibiting length variation in fragile X syndrome. *Cell* 65: 905-914.
2. Pieretti, M., et al. 1991. Absence of expression of the FMR-1 gene in fragile X syndrome. *Cell* 66: 817-22.
3. Zhang, Y., et al. 1995. The fragile X mental retardation syndrome protein interacts with novel homologs FXR1 and FXR2. *EMBO J.* 14: 5358-5366.
4. Deissler, H., et al. 1996. Purification of nuclear proteins from human HeLa cells that bind specifically to the unstable tandem repeat (CGG)_n in the human FMR1 gene. *J. Biol. Chem.* 271: 4327-4334.
5. Eberhart, D.E., et al. 1996. The fragile X mental retardation protein is a ribonucleoprotein containing both nuclear localization and nuclear export signals. *Hum. Mol. Genet.* 5: 1083-1091.

CHROMOSOMAL LOCATION

Genetic locus: Cggbp1 (mouse) mapping to 16 C1.3.

PRODUCT

CGGBP1 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 CGGBP1 shRNA Plasmid (m): sc-142304-SH and CGGBP1 shRNA (m) Lentiviral Particles: sc-142304-V as alternate gene silencing products.

For independent verification of CGGBP1 (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-142304A, sc-142304B and sc-142304C.

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 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

CGGBP1 siRNA (m) is recommended for the inhibition of CGGBP1 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

CGGBP1 (G-8): sc-376482 is recommended as a control antibody for monitoring of CGGBP1 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 CGGBP1 gene expression knockdown using RT-PCR Primer: CGGBP1 (m)-PR: sc-142304-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.