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FIR siRNA (m): sc-145186

BACKGROUND

Activation of FUSE, the far-upstream element, is required for the proper expression of the mammalian gene c-Myc. The binding of FBP (FUSE-binding protein) to FUSE is necessary for c-Myc expression. The FBP interacting repressor, FIR, binds to the central DNA-binding domain of FBP and can serve as an overriding negative regulator of c-Myc promoter activity. FIR interacts with the TFIID complex, which is a multifunctional, multisubunit RNA polymerase II transcription factor that interacts with several DNA-binding transactivators. FIR blocks activator-dependent, but not basal transcription through TFIID. FIR shares identity with seven in absentia (sia) binding protein 1. FIR is expressed in spleen, thymus, prostate, small intestine, colon, and peripheral blood leukocytes, and with relatively higher levels of expression in testis and ovary.

REFERENCES

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2. Duncan, R.D., Bazar, L., Michelotti, G., Tomonaga, T., Krutzsch, H., Avigan, M. and Levens, D. 1994. A sequence-specific, single strand binding protein activates the far upstream of c-Myc and defines a new DNA binding motif. *Genes Dev.* 8: 465-480.
3. Bazar, L., Harris, V., Sunitha, I., Hartmann, D., and Avigan, M. 1995. A transactivator of c-Myc is coordinately regulated with the proto-oncogene during cellular growth. *Oncogene* 10: 2229-2238.
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6. Liu, J., Liusheng, H., Collins, I., Ge, H., Libuitti, D., Li, J., Egly, J.M. and Levens, D. 2000. The FBP interacting repressor targets TFIID to inhibit activated transcription. *Mol. Cell* 5: 331-341.

CHROMOSOMAL LOCATION

Genetic locus: Puf60 (mouse) mapping to 15 D3.

PRODUCT

FIR 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 FIR shRNA Plasmid (m): sc-145186-SH and FIR shRNA (m) Lentiviral Particles: sc-145186-V as alternate gene silencing products.

For independent verification of FIR (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-145186A, sc-145186B and sc-145186C.

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

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

FIR (E-6): sc-398799 is recommended as a control antibody for monitoring of FIR 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 FIR gene expression knockdown using RT-PCR Primer: FIR (m)-PR: sc-145186-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.