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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

BRF2 siRNA (m): sc-141744

BACKGROUND

Human cells contain two types of RNA polymerase III transcription factor (TFIIIB), BRF1 and BRF2. BRF1, functions in recruitment of RNA polymerase III to the promoter for multiple rounds of transcription. BRF1 precisely positions TFIIIB on the upstream promoter-less DNA by creating stable protein-protein interactions with TATA-binding protein (TBP), another component of TFIIIB. BRF2, is recruited to type 3 promoters such as the human U6 snRNA promoter. It differs from BRF1-TFIIIB in that it contains the TFIIIB-related factor BRF2 instead of BRF1 and its three components do not form a stable complex.

REFERENCES

1. Wang, Z., et al. 1995. Structure and function of a human transcription factor TFIIIB subunit that is evolutionarily conserved and contains both TFIIIB- and high-mobility-group protein 2-related domains. *Proc. Natl. Acad. Sci. USA* 92: 7026-7030.
2. Kassavetis, G.A., et al. 1995. Cloning, expression, and function of TFC5, the gene encoding the B' component of the *Saccharomyces cerevisiae* RNA polymerase III transcription factor TFIIIB. *Proc. Natl. Acad. Sci. USA* 92: 9786-9790.
3. Whitehall, S.K., et al. 1995. The symmetry of the yeast U6 RNA gene's TATA box and the orientation of the TATA-binding protein in yeast TFIIIB. *Genes Dev.* 9: 2974-2985.
4. Ishiguro, A., et al. 2002. Essential roles of BDP1, a subunit of RNA polymerase III initiation factor TFIIIB, in transcription and tRNA processing. *Mol. Cell. Biol.* 22: 3264-3275.
5. Huang, Y., et al. 2003. The fission yeast TFIIIB-related factor limits RNA polymerase III to a TATA-dependent pathway of TBP recruitment. *Nucleic Acids Res.* 31: 2108-2116.
6. Hu, P., et al. 2004. CK2 phosphorylation of BDP1 executes cell cycle-specific RNA polymerase III transcription repression. *Mol. Cell* 16: 81-92.

CHROMOSOMAL LOCATION

Genetic locus: Brf2 (mouse) mapping to 8 A2.

PRODUCT

BRF2 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 BRF2 shRNA Plasmid (m): sc-141744-SH and BRF2 shRNA (m) Lentiviral Particles: sc-141744-V as alternate gene silencing products.

For independent verification of BRF2 (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-141744A, sc-141744B and sc-141744C.

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

BRF2 siRNA (m) is recommended for the inhibition of BRF2 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

BRF2 (C-8): sc-390312 is recommended as a control antibody for monitoring of BRF2 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 BRF2 gene expression knockdown using RT-PCR Primer: BRF2 (m)-PR: sc-141744-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.