SANTA CRUZ BIOTECHNOLOGY, INC.

NRF-1 siRNA (m): sc-38106

BACKGROUND

Nuclear respiratory factor-1 (NRF-1) is a transcriptional activator that has been implicated in the nuclear control of respiratory chain expression in mammalian cells. The NRF-1 gene is expressed during oogenesis and during the early stages of embryogenesis. *In vitro* studies have implicated NRF-1 in the transcriptional expression of nuclear genes required for mitochondrial respiratory function, as well as for other fundamental cellular activities. While most isolated wild-type and NRF-$1^{+/-}$ blastocysts continue to develop normally *in vitro,* NRF-$1^{-/-}$ blastocysts lack this ability, despite their normal morphology. NRF-1 is specifically required in the maintenance of mtDNA and respiratory chain function during early embryogenesis. NRF-1 also plays a key role in cellular adaptation to energy demands by translating physiological signals into an increased capacity for generating energy. Additionally, NRF-1 is a major transcription factor that binds the promoter in brain and testis.

REFERENCES

1. Huo, L. and Scarpulla, R.C. 1999. Multiple 5'-untranslated exons in the nuclear respiratory factor-1 gene span 47 kb and contribute to transcript heterogeneity and translational efficiency. Gene 233: 213-224.
2. Li, B., et al. 1999. Respiratory uncoupling induces δ-aminolevulinate synthase expression through a nuclear respiratory factor-1-dependent mechanism in HeLa cells. J. Biol. Chem. 25: 17534-17540.
3. Herzig, R.P., et al. 2000. Dynein light chain interacts with NRF-1 and EWG, structurally and functionally related transcription factors from humans and *Drosophila*. J. Cell Sci. 23: 4263-4273.
4. Huo, L. and Scarpulla, R.C. 2001. Mitochondrial DNA instability and peri-implantation lethality associated with targeted disruption of nuclear respiratory factor-1 in mice. Mol. Cell. Biol. 2: 644-654.
5. Kumari, D. and Usdin, K. 2001. Interaction of the transcription factors USF-1, USF-2 and α-Pal/NRF-1 with the FMR1 promoter. Implications for Fragile X mental retardation syndrome. J. Biol. Chem. 6: 4357-4364.

CHROMOSOMAL LOCATION

Genetic locus: Nrf1 (mouse) mapping to 6 A3.3.

PRODUCT

NRF-1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NRF-1 shRNA Plasmid (m): sc-38106-SH and NRF-1 shRNA (m) Lentiviral Particles: sc-38106-V as alternate gene silencing products.

For independent verification of NRF-1 (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-38106A, sc-38106B and sc-38106C.

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

NRF-1 siRNA (m) is recommended for the inhibition of NRF-1 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

NRF-1 (147.1): sc-101102 is recommended as a control antibody for monitoring of NRF-1 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 NRF-1 gene expression knockdown using RT-PCR Primer: NRF-1 (m)-PR: sc-38106-PR (20 µl, 588 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.