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Nrf1 siRNA (m): sc-43576

BACKGROUND

The NF-E2 DNA binding protein is composed of two subunits, p45 and MafK, and it regulates expression of globin genes in developing erythroid cells through interaction with Maf recognition elements (MAREs). A family of NF-E2-related proteins, which are collectively known as the cap "n" collar (CNC) family and include Nrf1 (also designated TCF11), Nrf2 and Nrf3, are bZIP transcription factors that heterodimerize with Maf proteins to bind MARE sequences. The Nrf proteins also bind the antioxidant response element (ARE) and are implicated in the regulation of detoxification enzymes and the oxidative stress response. They do so by heterodimerizing with Jun family members (c-Jun, Jun B and Jun D) to activate gene expression, specifically the detoxifying enzyme NQO1. The genes encoding Nrf1 and Nrf2 map to human chromosomes 17q21.32 and 2q31.2, respectively. Nrf2 is widely expressed and is thought to translocate to the nucleus after treatment with xenobiotics and antioxidants, which stimulate its release from a repressor protein Keap1. The gene encoding human Nrf3 maps to chromosome 7p15.2. Nrf3 is highly expressed in placenta, B cells and monocytes.

REFERENCES

- Chan, J.Y., et al. 1995. Chromosomal localization of the human NF-E2 family of bZIP transcription factors by fluorescence *in situ* hybridization. *Hum. Genet.* 95: 265-269.
- Chan, K., et al. 1996. Nrf2, a member of the NF-E2 family of transcription factors, is not essential for murine erythropoiesis, growth, and development. *Proc. Natl. Acad. Sci. USA* 93: 13943-13948.

CHROMOSOMAL LOCATION

Genetic locus: Nfe2l1 (mouse) mapping to 11 D.

PRODUCT

Nrf1 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 Nrf1 shRNA Plasmid (m): sc-43576-SH and Nrf1 shRNA (m) Lentiviral Particles: sc-43576-V as alternate gene silencing products.

For independent verification of Nrf1 (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-43576A, sc-43576B and sc-43576C.

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

Nrf1 siRNA (m) is recommended for the inhibition of Nrf1 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

Nrf1 (G-5): sc-515360 is recommended as a control antibody for monitoring of Nrf1 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 Nrf1 gene expression knockdown using RT-PCR Primer: Nrf1 (m)-PR: sc-43576-PR (20 μ l, 481 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Xing, W., et al. 2007. Nuclear factor-E2-related factor-1 mediates ascorbic acid induction of osterix expression via interaction with antioxidant-responsive element in bone cells. *J. Biol. Chem.* 282: 22052-22061.
- Zambrano, A., et al. 2014. The thyroid hormone receptor β induces DNA damage and premature senescence. *J. Cell Biol.* 204: 129-146.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

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