

Frataxin siRNA (h): sc-40580

BACKGROUND

Friedreich ataxia is a progressive neurodegenerative disorder caused by loss of function mutations in the Frataxin gene. The human Frataxin gene maps to chromosome 9q21.11. The Frataxin gene encodes a mitochondrial protein of the same name. Frataxin assembles into a stable homopolymer with iron-binding capabilities. When expressed in *E. coli*, human Frataxin binds iron atoms at a rate of ten iron atoms per one molecule of the Frataxin polymer. Thus, Frataxin appears to function in some capacity for iron-storage for the mitochondria. Frataxin may also function as an activator of oxidative phosphorylation to increase mitochondrial membrane potential and elevate cellular ATP. Frataxin is expressed in tissues with high metabolic activity including heart, liver and brown fat.

REFERENCES

1. Montermini, L., et al. 1995. The Friedreich ataxia critical region spans a 150-kb interval on chromosome 9q13. *Am. J. Hum. Genet.* 57: 1061-1067.
2. Campuzano, V., et al. 1997. Frataxin is reduced in Friedreich ataxia patients and is associated with mitochondrial membranes. *Hum. Mol. Genet.* 6: 1771-1780.
3. Koutnikova, H., et al. 1997. Studies of human, mouse and yeast homologues indicate a mitochondrial function for Frataxin. *Nat. Genet.* 16: 345-351.
4. Ristow, M., et al. 2000. Frataxin activates mitochondrial energy conversion and oxidative phosphorylation. *Proc. Natl. Acad. Sci. USA* 97: 12239-12243.
5. Cavadini, P., et al. 2002. Assembly and iron-binding properties of human Frataxin, the protein deficient in Friedreich ataxia. *Hum. Mol. Genet.* 11: 217-227.

CHROMOSOMAL LOCATION

Genetic locus: FXN (human) mapping to 9q21.11.

PRODUCT

Frataxin siRNA (h) 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 Frataxin shRNA Plasmid (h): sc-40580-SH and Frataxin shRNA (h) Lentiviral Particles: sc-40580-V as alternate gene silencing products.

For independent verification of Frataxin (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-40580A, sc-40580B and sc-40580C.

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

Frataxin siRNA (h) is recommended for the inhibition of Frataxin expression in human 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

Frataxin (G-12): sc-518079 is recommended as a control antibody for monitoring of Frataxin 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 Frataxin gene expression knockdown using RT-PCR Primer: Frataxin (h)-PR: sc-40580-PR (20 μ l, 492 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ohgari, Y., et al. 2011. Roles of porphyrin and iron metabolisms in the δ -aminolevulinic acid (ALA)-induced accumulation of protoporphyrin and photodamage of tumor cells. *Photochem. Photobiol.* 87: 1138-1145.

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.