

EF-G2 siRNA (h): sc-91850

BACKGROUND

Mitochondrial translation is critical for maintaining function and preventing impairment of mitochondrial DNA, with mitochondrial translation elongation factors playing an essential role in this process. EF-G2 (elongation factor G2), also known as RRF2mt (ribosome-releasing factor 2, mitochondrial), GFM2 or MSTP027, is a 779 amino acid protein belonging to the GTP-binding elongation factor family and EF-G/EF-2 subfamily. Localizing to mitochondrion, EF-G2 is widely expressed with high levels found in adult heart, skeletal muscle and kidney as well as adult and fetal liver. EF-G2 mediates ribosome recycling and by interacting directly with MRRF on the ribosome large subunit, is required for ribosome dissociation from mRNA. Existing as five alternatively spliced isoforms, the gene encoding EF-G2 maps to human chromosome 5q13.3.

REFERENCES

1. Hammarsund, M., et al. 2001. Identification and characterization of two novel human mitochondrial elongation factor genes, hEFG2 and hEFG1, phylogenetically conserved through evolution. *Hum. Genet.* 109: 542-550.
2. Antonicka, H., et al. 2006. The molecular basis for tissue specificity of the oxidative phosphorylation deficiencies in patients with mutations in the mitochondrial translation factor EFG1. *Hum. Mol. Genet.* 15: 1835-1846.
3. Li, J., et al. 2007. Separation and identification of the exosomes derived from a mouse hepatoma carcinoma cell line (H22) and initial investigation of their protein composition. *Zhonghua Gan Zang Bing Za Zhi* 15: 437-440.
4. Sasarman, F., et al. 2008. The A3243G tRNA^{Leu}(UUR) MELAS mutation causes amino acid misincorporation and a combined respiratory chain assembly defect partially suppressed by overexpression of EFTu and EFG2. *Hum. Mol. Genet.* 17: 3697-3707.
5. Christian, B., et al. 2009. Ribosome shifting or splitting: it is all up to the EF-G. *Mol. Cell* 35: 400-402.
6. Tsuboi, M., et al. 2009. EF-G2mt is an exclusive recycling factor in mammalian mitochondrial protein synthesis. *Mol. Cell* 35: 502-510.
7. Suematsu, T., et al. 2010. A bacterial elongation factor G homologue exclusively functions in ribosome recycling in the spirochaete *Borrelia burgdorferi*. *Mol. Microbiol.* 75: 1445-1454.

CHROMOSOMAL LOCATION

Genetic locus: GFM2 (human) mapping to 5q13.3.

PRODUCT

EF-G2 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 EF-G2 shRNA Plasmid (h): sc-91850-SH and EF-G2 shRNA (h) Lentiviral Particles: sc-91850-V as alternate gene silencing products.

For independent verification of EF-G2 (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-91850A, sc-91850B and sc-91850C.

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

EF-G2 siRNA (h) is recommended for the inhibition of EF-G2 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

EF-G2 (E-10): sc-514242 is recommended as a control antibody for monitoring of EF-G2 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 EF-G2 gene expression knockdown using RT-PCR Primer: EF-G2 (h)-PR: sc-91850-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.

PROTOCOLS

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