

FT α siRNA (h): sc-35420

BACKGROUND

Mammalian protein farnesyl transferases are heterodimeric proteins containing two nonidentical α and β subunits that attach farnesyl residues to a cysteine at the fourth position from the COOH terminus of several proteins, including nuclear lamins and p21Ras proteins. The natural substrates contain the Cys-A-A-Xaa recognition sequence, where the A residues are aliphatic and Xaa represents methionine, serine, glutamine or cysteine. The purified farnesyl transferase is an $\alpha\beta$ heterodimer. The β subunit binds the peptide substrate while the α subunit is suspected to participate in formation of a stable complex with the substrate farnesyl pyrophosphate. The α subunit is shared with a second prenyltransferase, geranylgeranyl transferase, that attaches 20 carbon geranylgeranyl to Ras-related proteins that terminate in a Cys-A-A-Xaa recognition site in which Xaa is leucine.

REFERENCES

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2. Reiss, Y., et al. 1990. Inhibition of purified p21Ras farnesyl: protein transferase by Cys-AAX tetrapeptides. *Cell* 62: 81-88.
3. Reiss, Y., et al. 1991. Sequence requirement for peptide recognition by rat brain p21Ras protein farnesyltransferase. *Proc. Natl. Acad. Sci. USA* 88: 732-736.
4. Chen, W.J., et al. 1991. Cloning and expression of a cDNA encoding the α subunit of rat p21Ras protein farnesyltransferase. *Proc. Natl. Acad. Sci. USA* 88: 11368-11372.
5. Reiss, Y., et al. 1991. Nonidentical subunits of p21H-Ras farnesyltransferase. *J. Biol. Chem.* 266: 10672-10677.
6. Moores, S.L., et al. 1991. Sequence dependence of protein isoprenylation. *J. Biol. Chem.* 266: 14603-14610.
7. Seabra, M.C., et al. 1991. Protein farnesyltransferase and geranylgeranyltransferase share a common α subunit. *Cell* 65: 429-434.
8. Andres, D.A., et al. 1993. cDNA cloning of the two subunits of human CAAX farnesyltransferase and chromosomal mapping of FNTA and FNTB loci and related sequences. *Genomics* 18: 105-112.

CHROMOSOMAL LOCATION

Genetic locus: FTNA (human) mapping to 8p11.21.

PRODUCT

FT α 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 FT α shRNA Plasmid (h): sc-35420-SH and FT α shRNA (h) Lentiviral Particles: sc-35420-V as alternate gene silencing products.

For independent verification of FT α (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-35420A, sc-35420B and sc-35420C.

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

FT α siRNA (h) is recommended for the inhibition of FT α 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

FT α (D-5): sc-374262 is recommended as a control antibody for monitoring of FT α 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 FT α gene expression knockdown using RT-PCR Primer: FT α (h)-PR: sc-35420-PR (20 μ l, 597 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.

PROTOCOLS

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