SANTA CRUZ BIOTECHNOLOGY, INC.

FTβ siRNA (r): sc-77354



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 α - β heterodimer. The β subunit, which is known as FT β , CAAX farnesyltransferase subunit β , or Ras proteins prenyltransferase subunit β , is a 437 amino acid protein that contains five PFTB repeats and binds the peptide substrate. The α subunit is suspected to participate in formation of a stable complex with the substrate farnesyl pyrophosphate.

REFERENCES

- Clarke, S., et al. 1988. Posttranslational modification of the Ha-Ras oncogene protein: evidence for a third class of protein carboxyl methyltransferases. Proc. Natl. Acad. Sci. USA 85: 4643-4647.
- Reiss, Y., et al.1990. Inhibition of purified p21Ras farnesyl: protein transferase by Cys-AAX tetrapeptides. Cell 62: 81-88.
- 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. Moores, S.L., et al. 1991. Sequence dependence of protein isoprenylation. J. Biol. Chem 266: 14603-14610.
- 5. Seabra, M.C., et al. 1991. Protein farnesyltransferase and geranylgeranyltransferase share a common a subunit. Cell 65: 429-434.
- Reiss, Y., et al. 1991. Nonidentical subunits of p21H-Ras farnesyltransferase. J. Biol. Chem. 266: 10672-10677.
- Chen, W.J., et al.1991. Cloning and expression of a cDNA encoding the a subunit of rat p21Ras protein farnesyltransferase. Proc. Natl. Acad. Sci. USA 88: 11368-11372.

CHROMOSOMAL LOCATION

Genetic locus: Fntb (rat) mapping to 6q24.

PRODUCT

FT β siRNA (r) 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 β shRNA Plasmid (r): sc-77354-SH and FT β shRNA (r) Lentiviral Particles: sc-77354-V as alternate gene silencing products.

For independent verification of FT β (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-77354A, sc-77354B and sc-77354C.

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 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 β siRNA (r) is recommended for the inhibition of FT β expression in rat 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 β (B-7): sc-46664 is recommended as a control antibody for monitoring of FT β 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 MarkerTM 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 β gene expression knockdown using RT-PCR Primer: FT β (r)-PR: sc-77354-PR (20 µl). 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.