GGTase-Iβ siRNA (h): sc-40882



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BACKGROUND

Eukaryotic cells contain three distinct prenyltransferases that catalyze the attachment of a thioether-linked 15-carbon farnesyl group or 20-carbon geranylgeranyl group to C-terminal cysteine residues. Geranylgeranyltransferase type I (GGTase-I, PGGTase-I) catalyzes the nucleophilic substitution reaction between geranylgeranyl diphosphate (GGPP) and a protein-derived thiol to form the thioether linkage. The candidate protein contains a C-terminal CAAX motif in which "A" is an aliphatic amino acid and "X" is leucine. Geranylgeranylation is necessary for the TGF β 1 signaling pathway, which involves phosphatidylcholine-specific phospholipase and a protein kinase C. Human GGTase-I contains an α subunit and a β subunit. Geranylgeranyltransferase type II (GGTase-II) is a heterodimer that catalyzes the transfer of two 20-carbon geranylgeranyl groups from geranylgeranyl pyrophosphate onto C-terminal cysteine residues of Rab GTPases, which is required for the activity of Rab proteins. GGTase-II also contains an α subunit and a β subunit.

REFERENCES

- 1. Schafer, W.R., et al. 1992. Protein prenylation: genes, enzymes, targets, and functions. Annu. Rev. Genet. 26: 209-237.
- 2. van Bokhoven, H., et al. 1996. cDNA cloning and chromosomal localization of the genes encoding the α and β subunits of human Rab geranylgeranyl transferase: the 3' end of the α subunit gene overlaps with the transglutaminase 1 gene promoter. Genomics 38: 133-140.
- 3. Online Mendelian Inheritance in Man, OMIM™. 1997. Johns Hopkins University, Baltimore, MD. MIM Number: 602031. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/

CHROMOSOMAL LOCATION

Genetic locus: PGGT1B (human) mapping to 5q22.3.

PRODUCT

GGTase-I β 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 GGTase-I β shRNA Plasmid (h): sc-40882-SH and GGTase-I β shRNA (h) Lentiviral Particles: sc-40882-V as alternate gene silencing products.

For independent verification of GGTase-I β (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-40882A, sc-40882B and sc-40882C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20 $^{\circ}$ C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20 $^{\circ}$ 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

GGTase-I β siRNA (h) is recommended for the inhibition of GGTase-I β 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 µM in 66 µl. 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

GGTase-Iβ (D-11): sc-376854 is recommended as a control antibody for monitoring of GGTase-Iβ 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-lgG κ BP-HRP: sc-516102 or m-lgG 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-lgG κ BP-FITC: sc-516140 or m-lgG 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 GGTase-I β gene expression knockdown using RT-PCR Primer: GGTase-I β (h)-PR: sc-40882-PR (20 μ I, 575 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

 Virtanen, S.S., et al. 2010. Inhibition of GGTase-I and FTase disrupts cytoskeletal organization of human PC-3 prostate cancer cells. Cell Biol. Int. 34: 815-826.

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.

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