SANTA CRUZ BIOTECHNOLOGY, INC.

SnoA/N siRNA (m): sc-36519

BACKGROUND

The Ski family of oncogenes includes Ski, SnoN and SnoA, which are produced by alternative splicing of the Sno gene. Ski family members are nuclear proteins that form homodimers and heterodimers, bind to DNA and function as transcriptional activators and repressors. These proteins consist of five tandem repeats in the C-terminal domain and two leucine zipper motifs that are responsible for efficient DNA binding, trimerization and cellular transformation. The Ski proteins regulate TGFβ induced gene-specific transcriptional activation by effectively repressing Smad activity and, thereby, inhibit TGFβ induced cell growth and extracellular matrix production. The amino terminus of Ski and SnoN preferentially associate with the MH2 domain of Smad2 and Smad4 of the Smad family of transcription factors, where they then recruit the transcriptional co-repressor protein N-CoR to the complex to inhibit transcription. Alternatively, Ski proteins are negatively regulated by various Smad proteins, as TGFβ induces Smad3 accumulation in the nucleus, where it is then responsible for inducing the rapid degradation of SnoN and faciliating TGFβ signaling pathways and Smad-activated gene transcription.

REFERENCES

1. Nomura, N., et al. 1989. Isolation of human cDNA clones of Ski and the Ski-related gene, Sno. Nucleic Acids Res. 17: 5489-5500.
2. Pearson-White, S. 1993. SnoL, a novel alternatively spliced isoform of the Ski proto-oncogene homolog, Sno. Nucleic Acids Res. 21: 4632-4638.
3. Nagase, T., et al. 1993. Complex formation between proteins encoded by the Ski gene family. J. Biol. Chem. 268: 13710-13716.

CHROMOSOMAL LOCATION

Genetic locus: Skil (mouse) mapping to 3 A3.

PRODUCT

SnoA/N siRNA (m) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see SnoA/N shRNA Plasmid (m): sc-36519-SH and SnoA/N shRNA (m) Lentiviral Particles: sc-36519-V as alternate gene silencing products.

For independent verification of SnoA/N (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-36519A, sc-36519B and sc-36519C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

SnoA/N siRNA (m) is recommended for the inhibition of SnoA/N expression in mouse 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

SnoN (B-3): sc-136958 is recommended as a control antibody for monitoring of SnoA/N 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 SnoA/N gene expression knockdown using RT-PCR Primer: SnoA/N (m)-PR: sc-36519-PR (20 µl, 472 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Mistry, D.S., et al. 2011. Gonadotropin-releasing hormone pulse sensitivity of follicle-stimulating hormone-β gene is mediated by differential expression of positive regulatory activator protein 1 factors and corepressors SKIL and TGIF1. Mol. Endocrinol. 25: 1387-1403.

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.