

SIRT5 siRNA (h): sc-63026

BACKGROUND

Sirtuins (SIRT1-7) are human homologs of the yeast Sir2 (silent information regulator-2) protein and are divided into four main classes: SIRT1-3 are class I, SIRT4 is class II, SIRT5 is class III and SIRT6-7 are class IV. In *S. cerevisiae*, Sir2 deacetylates histones in an NAD-dependent manner, which regulates silencing at the telomeric, rDNA (ribosomal RNA) and silent mating-type loci. The human SIRT proteins are NAD-dependent deacetylases that act as intracellular regulators and are thought to have ribosyltransferase activity. SIRT5 (NAD-dependent deacetylase sirtuin-5), also known as SIR2L5, is a 310 amino acid member of the class III sirtuins. Localized to mitochondria and expressed throughout the body, SIRT5 is an NAD-dependent deacetylase that may link metabolic aging processes in humans. SIRT5 contains one deacetylase-sirtuin-type domain and can be deactivated by suramin, a drug that blocks the binding of various growth factors. Two isoforms of SIRT5 exist due to alternative splicing events.

REFERENCES

1. Frye, R.A. 1999. Characterization of five human cDNAs with homology to the yeast SIR2 gene: Sir2-like proteins (sirtuins) metabolize NAD and may have protein ADP-ribosyltransferase activity. *Biochem. Biophys. Res. Commun.* 260: 273-279.
2. Frye, R.A. 2000. Phylogenetic classification of prokaryotic and eukaryotic Sir2-like proteins. *Biochem. Biophys. Res. Commun.* 273: 793-798.
3. Kyrylenko, S., et al. 2003. Differential regulation of the Sir2 histone deacetylase gene family by inhibitors of class I and II histone deacetylases. *Cell. Mol. Life Sci.* 60: 1990-1997.

CHROMOSOMAL LOCATION

Genetic locus: SIRT5 (human) mapping to 6p23.

PRODUCT

SIRT5 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 SIRT5 shRNA Plasmid (h): sc-63026-SH and SIRT5 shRNA (h) Lentiviral Particles: sc-63026-V as alternate gene silencing products.

For independent verification of SIRT5 (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-63026A, sc-63026B and sc-63026C.

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

SIRT5 siRNA (h) is recommended for the inhibition of SIRT5 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

SIRT5 (G-2): sc-271635 is recommended as a control antibody for monitoring of SIRT5 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 SIRT5 gene expression knockdown using RT-PCR Primer: SIRT5 (h)-PR: sc-63026-PR (20 μ l, 443 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Li, D.J., et al. 2021. NAD⁺-boosting therapy alleviates nonalcoholic fatty liver disease via stimulating a novel exerkine Fndc5/irisin. *Theranostics* 11: 4381-4402.
2. Wang, Y., et al. 2022. SIRT4-catalyzed deacetylation of Axin1 modulates the Wnt/ β -catenin signaling pathway. *Front. Oncol.* 12: 872444.

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.