

Nucling siRNA (h): sc-90147

BACKGROUND

Nucling, also known as UACA (uveal autoantigen with coiled-coil domains and ankyrin repeats) and KIAA1561, is a 1,416 amino acid nuclear and cytoplasmic protein. Upregulated after TSH-stimulation, Nucling is a component of the apoptosome complex, whose other components include Apaf-1 and caspase-9. Nucling interacts directly with Apaf-1 and regulates its redistribution to the nucleus following proapoptotic stress. Nucling also plays a role in the promotion of apoptosis by the galectin-3 downregulation, apoptosome upregulation and NF κ B inactivation pathways. Nucling also interacts with ARF6, which may modulate cell shape and motility following injury. Nucling contains six ANK repeats and is expressed highly in kidney, heart, pancreas and skeletal muscle. Nucling is a potential target autoantigen in Behcet disease (BD), Vogt-Koyanagi-Harada (VKH) and sarcoidosis, which cause different types of panuveitis.

REFERENCES

1. Yamada, K., et al. 2001. Identification of a novel autoantigen UACA in patients with panuveitis. *Biochem. Biophys. Res. Commun.* 280: 1169-1176.
2. Ohkura, T., et al. 2004. Detection of the novel autoantibody (anti-UACA antibody) in patients with Graves' disease. *Biochem. Biophys. Res. Commun.* 321: 432-440.
3. Brandenberger, R., et al. 2004. Transcriptome characterization elucidates signaling networks that control human ES cell growth and differentiation. *Nat. Biotechnol.* 22: 707-716.
4. Bouwmeester, T., et al. 2004. A physical and functional map of the human TNF α /NF κ B signal transduction pathway. *Nat. Cell Biol.* 6: 97-105.
5. Beausoleil, S.A., et al. 2004. Large-scale characterization of HeLa cell nuclear phosphoproteins. *Proc. Natl. Acad. Sci. USA* 101: 12130-12135.
6. Trynka, G., et al. 2009. Coeliac disease-associated risk variants in TNFAIP3 and REL implicate altered NF κ B signalling. *Gut* 58: 1078-1083.
7. Choudhary, C., et al. 2009. Lysine acetylation targets protein complexes and co-regulates major cellular functions. *Science* 325: 834-840.

CHROMOSOMAL LOCATION

Genetic locus: UACA (human) mapping to 15q23.

PRODUCT

Nucling 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 Nucling shRNA Plasmid (h): sc-90147-SH and Nucling shRNA (h) Lentiviral Particles: sc-90147-V as alternate gene silencing products.

For independent verification of Nucling (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-90147A, sc-90147B and sc-90147C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

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

Nucling (A-5): sc-515005 is recommended as a control antibody for monitoring of Nucling 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 Nucling gene expression knockdown using RT-PCR Primer: Nucling (h)-PR: sc-90147-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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