



Artemis siRNA (h): sc-42734

BACKGROUND

Distinct DNA repair pathways minimize the consequences of mutagenic events. Reactive oxygen species (ROS) are highly reactive atoms with an unpaired electron that are conducive to double-strand DNA breaking events. Artemis, named after the Greek goddess for the protection of children, is one of the major proteins contributing to the preservation of double-strand breaks in DNA by cutting away the damaged parts of the DNA, which allows the strands to rejoin. Artemis is a single-strand-specific 5' to 3' exonuclease that forms a complex with the DNA-dependent protein kinase (DNA-PK_{CS}). DNA-PK_{CS} phosphorylates Artemis, and Artemis acquires endonucleolytic activity on 5' and 3' overhangs and hairpins. These activities are essential for V(D)J recombination and for the 5' and 3' overhang processing in non-homologous DNA end joining. Mutations in the human Artemis protein result in hypersensitivity to DNA double-strand break-inducing agents and absence of B and T lymphocytes, which is known as "bubble boy" disease or severe combined immunodeficiency disease (SCID). The human Artemis gene maps to chromosome 10p13, and encodes a 577 amino acid protein.

REFERENCES

1. Li, L., et al. 1998. The gene for severe combined immunodeficiency disease in Athabascan-speaking Native Americans is located on chromosome 10p. *Am. J. Hum. Genet.* 62: 136-144.
2. Moshous, D., et al. 2001. Artemis, a novel DNA double-strand break repair/V(D)J recombination protein, is mutated in human severe combined immune deficiency. *Cell* 105: 177-186.
3. Wood, R.D., et al. 2001. Human DNA repair genes. *Science* 291: 1284-1289.

CHROMOSOMAL LOCATION

Genetic locus: DCLRE1C (human) mapping to 10p13.

PRODUCT

Artemis 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 Artemis shRNA Plasmid (h): sc-42734-SH and Artemis shRNA (h) Lentiviral Particles: sc-42734-V as alternate gene silencing products.

For independent verification of Artemis (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-42734A, sc-42734B and sc-42734C.

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

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

Artemis (C-8): sc-518193 is recommended as a control antibody for monitoring of Artemis 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 Artemis gene expression knockdown using RT-PCR Primer: Artemis (h)-PR: sc-42734-PR (20 μ l, 432 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Arthur, L.L., et al. 2017. Rapid generation of hypomorphic mutations. *Nat. Commun.* 8: 14112.
2. Munnur, D., et al. 2019. NR4A nuclear receptors target poly-ADP-ribosylated DNA-PK_{CS} protein to promote DNA repair. *Cell Rep.* 26: 2028-2036.e6.

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.