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ARD1 siRNA (m): sc-44714

BACKGROUND

The ARD1 subfamily of proteins belongs to the larger acetyltransferase family. N-terminal acetyltransferase complex ARD1, also designated Te2, forms a complex with NARG1, displaying N-terminal acetyltransferase activity. Without NARG1, ARD1 promotes hypoxia-inducible factor-1 α (HIF-1 α) degradation by displaying internal acetyltransferase activity towards HIF-1 α . This ubiquitously expressed protein, which is mainly cytoplasmic, is cleaved by caspases during apoptosis. ARD1 interacts with the ribosome, NARG1 and HIF-1 α . In its binding to HIF-1 α , ARD1 acts as a protein acetyltransferase by regulating its stability. In many cell lines, ARD1 is downregulated in response to hypoxia. ARD1 is expressed throughout the developing brain.

REFERENCES

1. Jeong, J.W., et al. 2002. Regulation and destabilization of HIF-1 α by ARD1-mediated acetylation. *Cell* 111: 709-720.
2. Sugiura, N., et al. 2003. An evolutionarily conserved N-terminal acetyltransferase complex associated with neuronal development. *J. Biol. Chem.* 278: 40113-40120.
3. Carninci, P., et al. 2005. The transcriptional landscape of the mammalian genome. *Science* 309: 1559-1563.
4. Fisher, T.S., et al. 2005. Analysis of ARD1 function in hypoxia response using retroviral RNA interference. *J. Biol. Chem.* 280: 17749-17757.
5. Vichi, A., et al. 2005. E3 ubiquitin ligase activity of the trifunctional ARD1 (ADP-ribosylation factor domain protein 1). *Proc. Natl. Acad. Sci. USA* 102: 1945-1950.
6. Lee, K.H., et al. 2006. Differential responses of two degradation domains of HIF-1 α to hypoxia and iron deficiency. *Biochimie* 88: 163-169.

CHROMOSOMAL LOCATION

Genetic locus: Naa10 (mouse) mapping to X A7.3.

PRODUCT

ARD1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ARD1 shRNA Plasmid (m): sc-44714-SH and ARD1 shRNA (m) Lentiviral Particles: sc-44714-V as alternate gene silencing products.

For independent verification of ARD1 (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-44714A, sc-44714B and sc-44714C.

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.

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

ARD1 siRNA (m) is recommended for the inhibition of ARD1 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 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

ARD1 (A-10): sc-373920 is recommended as a control antibody for monitoring of ARD1 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 ARD1 gene expression knockdown using RT-PCR Primer: ARD1 (m)-PR: sc-44714-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.