

Arkadia siRNA (h): sc-40346

BACKGROUND

Communication between cells during early embryogenesis establishes the basic organization of the vertebrate body plan. The early mammalian embryo is patterned by signals emanating from extraembryonic and embryonic signaling centers, most notably the anterior visceral endoderm (AVE) and the node, respectively. Nodal-related members of the transforming growth factor (TGF)- β family regulate the induction of mesoderm, endoderm and mesoderm. The different tissues form in response to the same signaling molecules, which may elicit differential responses through concentration-dependent effects, extracellular cofactors and antagonists, or factor potentiation. The nuclear protein Arkadia specifically potentiates the mesendoderm-inducing activity of a subset of TGF- β family members and interacts with Nodal, which mediates the function of Arkadia in node induction. Arkadia functions within extraembryonic tissues and is required to induce the node as an essential modulator of the nodal signalling cascade. The human gene for Arkadia maps to chromosome 15q22.1 and encodes a 441 amino acid protein. The murine gene for arkadia maps to chromosome 9 39.0 cM and encodes a 989 amino acid protein.

REFERENCES

- Schier, A.F. and Shen, M.M. 2000. Nodal signalling in vertebrate development. *Nature* 403: 385-389.
- Episkopou, V., Arkell, R., Timmons, P.M., Walsh, J.J., Andrew, R.L. and Swan, D. 2001. Induction of the mammalian node requires Arkadia function in the extraembryonic lineages. *Nature* 410: 825-830.
- Niederlander, C., Walsh, J.J., Episkopou, V. and Jones, C.M. 2001. Arkadia enhances nodal-related signalling to induce mesendoderm. *Nature* 410: 830-834.
- Patten, I. and Placzek, M. 2001. Vertebrate development: Et in Arkadia. *Curr. Biol.* 11: 616-619.
- LocusLink Report (LocusID: 54778). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: RNF111 (human) mapping to 15q22.1.

PRODUCT

Arkadia 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 Arkadia shRNA Plasmid (h): sc-40346-SH and Arkadia shRNA (h) Lentiviral Particles: sc-40346-V as alternate gene silencing products.

For independent verification of Arkadia (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-40346A, sc-40346B and sc-40346C.

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

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

Arkadia (1-KK14): sc-134270 is recommended as a control antibody for monitoring of Arkadia 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 Arkadia gene expression knockdown using RT-PCR Primer: Arkadia (h)-PR: sc-40346-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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