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Lfc siRNA (h): sc-41723

BACKGROUND

Guanine nucleotide exchange factors (GEFs) regulate the activity of the small GTP-binding proteins of the Ras superfamily. The Ras family of proteins participate in a diverse array of functions, including signal transduction, cytoskeleton dynamics and intracellular trafficking. Ras proteins cycle between a biologically inactive GDP-bound state and an active GTP-bound state. GEFs catalyze the release of GDP from inactive Ras proteins to enable GTP binding. Lfc (also designated Rho/Rac guanine nucleotide exchange factor 2, lymphoid blast crisis-like 1, ARHGEF2, GEF-H1, KIAA0651 or LFP40) is an 894 amino acid GEF for Rho A, a member of the Ras family that regulates the Actin cytoskeleton during the formation of stress fibers. Lfc directly interacts with Cingulin, a junctional adapter that inhibits Rho A, thus preventing phorbol-12-myristate-13-acetate-induced contraction and apoptosis. Mutations in the p53 gene transcriptionally activate Lfc, thereby increasing tumor cell proliferation.

REFERENCES

1. Krendel, M., et al. 2002. Nucleotide exchange factor GEF-H1 mediates cross-talk between microtubules and the Actin cytoskeleton. *Nat. Cell Biol.* 4: 294-301.
2. Brecht, M., et al. 2004. Activation of GEF-H1, a guanine nucleotide exchange factor for Rho A, by DNA transfection. *Int. J. Cancer* 113: 533-540.
3. Zenke, F.T., et al. 2004. p21-activated kinase 1 phosphorylates and regulates 14-3-3 binding to GEF-H1, a microtubule-localized Rho exchange factor. *J. Biol. Chem.* 279: 18392-18400.
4. Aijaz, S., et al. 2005. Binding of GEF-H1 in inhibition of Rho signaling and G₁/S phase transition. *Dev. Cell* 8: 777-786.
5. Callow, M.G., et al. 2005. PAK4 mediates morphological changes through the regulation of GEF-H1. *J. Cell Sci.* 118: 1861-1872.
6. Online Mendelian Inheritance in Man, OMIM™. 2005. Johns Hopkins University, Baltimore, MD. MIM Number: 607560. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
7. Chang, Z.F. and Lee, H.H. 2006. Rho A signaling in phorbol ester-induced apoptosis. *J. Biomed. Sci.* 13: 173-180.

CHROMOSOMAL LOCATION

Genetic locus: ARHGEF2 (human) mapping to 1q22.

PRODUCT

Lfc siRNA (h) is a pool of 2 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 Lfc shRNA Plasmid (h): sc-41723-SH and Lfc shRNA (h) Lentiviral Particles: sc-41723-V as alternate gene silencing products.

For independent verification of Lfc (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-41723A and sc-41723B.

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

Lfc siRNA (h) is recommended for the inhibition of Lfc 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Lfc gene expression knockdown using RT-PCR Primer: Lfc (h)-PR: sc-41723-PR (20 μ l, 479 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Fukazawa, A., et al. 2008. GEF-H1 mediated control of NOD1 dependent NF κ B activation by Shigella effectors. *PLoS Pathog.* 4: e1000228.

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.