

ARF3 siRNA (h): sc-156161

BACKGROUND

The ADP-ribosylation factor (ARF) protein family are structurally and functionally conserved members of the Ras superfamily of regulatory GTP-binding proteins. ARFs influence vesicle trafficking and signal transduction in eukaryotic cells. ARF-dependent regulatory mechanisms include the coordination of spectrin interactions with Golgi membranes and the association of Actin to the Golgi via Rho family-dependent G protein localization and WASP/Arp2/3 complexes. Additionally, ARFs play a central role in the maintenance of organelle integrity, assembly of coat proteins and activation of phospholipase D (PC-PLD). ARF3 (ADP-ribosylation factor 3), is a 181 amino acid protein that localizes to Golgi apparatus and belongs to the small GTPase superfamily. ARF3 interacts with PICK1 and may modulate budding and uncoating of vesicles within the Golgi apparatus. Known to activate cholera toxin, ARF3 maps to human chromosome 12q13.12.

REFERENCES

1. Tsai, S.C., et al. 1991. Isolation and characterization of the human gene for ADP-ribosylation factor 3, a 20-kDa guanine nucleotide-binding protein activator of cholera toxin. *J. Biol. Chem.* 266: 23053-23059.
2. Hirai, M., et al. 1996. Assignment of human ADP ribosylation factor (ARF) genes ARF1 and ARF3 to chromosomes 1q42 and 12q13, respectively. *Genomics* 34: 263-265.
3. Hosaka, M., et al. 1996. Structure and intracellular localization of mouse ADP-ribosylation factors type 1 to type 6 (ARF1-ARF6). *J. Biochem.* 120: 813-819.
4. Boman, A.L., et al. 2000. A family of ADP-ribosylation factor effectors that can alter membrane transport through the *trans*-Golgi. *Mol. Biol. Cell* 11: 1241-1255.
5. Online Mendelian Inheritance in Man, OMIM[™]. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 103190. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
6. Irobi, J., et al. 2002. Mutation analysis of 12 candidate genes for distal hereditary motor neuropathy type II (distal HMN II) linked to 12q24.3. *J. Peripher. Nerv. Syst.* 7: 87-95.

CHROMOSOMAL LOCATION

Genetic locus: ARF3 (human) mapping to 12q13.12.

PRODUCT

ARF3 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 ARF3 shRNA Plasmid (h): sc-156161-SH and ARF3 shRNA (h) Lentiviral Particles: sc-156161-V as alternate gene silencing products.

For independent verification of ARF3 (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-156161A, sc-156161B and sc-156161C.

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

ARF3 siRNA (h) is recommended for the inhibition of ARF3 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

ARF3 (41): sc-135841 is recommended as a control antibody for monitoring of ARF3 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 ARF3 gene expression knockdown using RT-PCR Primer: ARF3 (h)-PR: sc-156161-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.

PROTOCOLS

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