

ARL4 siRNA (h): sc-61992

BACKGROUND

ADP-ribosylation factors (ARFs) are highly conserved guanine nucleotide-binding proteins that enhance the ADP-ribosyltransferase activity of cholera toxin. ARFs are important in eukaryotic vesicular trafficking pathways and activating phospholipase D. ARL4 (ADP-ribosylation factor-like protein 4A) is a member of the ARF-like protein (ARL) subfamily of small GTPases. It contains a C-terminal nuclear localization signal (NLS) region that interacts with Importin- α . ARL4 localizes to the nucleus and is found in a variety of tissues, but is predominantly expressed in spermatogonia and Sertoli cells. It is most closely related to ARL6 and ARL7. Unlike ARFs, ARL4 does not activate the cholera toxin ADP-ribosyltransferase. ARL4 may play a role in neurogenesis during embryonic development and somitogenesis in the early stages of adult spermatogenesis.

REFERENCES

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- Pasqualato, S., et al. 2002. Arf, Arl, Arp and Sar proteins: a family of GTP-binding proteins with a structural device for "front-back" communication. *EMBO Rep.* 3: 1035-1041.
- Schürmann, A., et al. 2002. Reduced sperm count and normal fertility in male mice with targeted disruption of the ADP-ribosylation factor-like 4 (Arl4) gene. *Mol. Cell. Biol.* 22: 2761-2768.

CHROMOSOMAL LOCATION

Genetic locus: ARL4A (human) mapping to 7p21.3.

PRODUCT

ARL4 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 ARL4 shRNA Plasmid (h): sc-61992-SH and ARL4 shRNA (h) Lentiviral Particles: sc-61992-V as alternate gene silencing products.

For independent verification of ARL4 (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-61992A, sc-61992B and sc-61992C.

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

ARL4 siRNA (h) is recommended for the inhibition of ARL4 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

ARL4 (B-10): sc-398352 is recommended as a control antibody for monitoring of ARL4 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 ARL4 gene expression knockdown using RT-PCR Primer: ARL4 (h)-PR: sc-61992-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.