



ARL8A siRNA (h): sc-88429

BACKGROUND

ADP-ribosylation factors (ARFs) are highly conserved guanine nucleotide binding proteins that enhance the ADP-ribosyltransferase activity of Cholera Toxin. ARF's are important in eukaryotic vesicular trafficking pathways and they play an essential role in the activation of phospholipase D (PC-PLD). ARL8A (ADP-ribosylation factor-like protein 8A), also known as ARL10B or GIE2, is a 186 amino acid protein that localizes to the membrane of late endosomes. Expressed ubiquitously, ARL8A is thought to play a role in lysosomal motility and may also be involved in chromosomal segregation events. The gene encoding ARL8A maps to human chromosome 1, which spans 260 million base pairs, houses over 3,000 genes and comprises nearly 8% of the human genome.

REFERENCES

1. Bonaldo, M.F., et al. 1996. Normalization and subtraction: two approaches to facilitate gene discovery. *Genome Res.* 6: 791-806.
2. Secombe, J., et al. 2004. *Drosophila* Topors is a RING finger-containing protein that functions as a ubiquitin-protein isopeptide ligase for the hairy basic helix-loop-helix repressor protein. *J. Biol. Chem.* 279: 17126-17133.
3. Okai, T., et al. 2004. Novel small GTPase subfamily capable of associating with tubulin is required for chromosome segregation. *J. Cell Sci.* 117: 4705-4715.
4. Hofmann, I., et al. 2006. An N-terminally acetylated Arf-like GTPase is localised to lysosomes and affects their motility. *J. Cell Sci.* 119: 1494-1503.
5. Burguete, A.S., et al. 2008. Rab and Arl GTPase family members cooperate in the localization of the golgin GCC185. *Cell* 132: 286-298.

CHROMOSOMAL LOCATION

Genetic locus: ARL8A (human) mapping to 1q32.1.

PRODUCT

ARL8A 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 ARL8A shRNA Plasmid (h): sc-88429-SH and ARL8A shRNA (h) Lentiviral Particles: sc-88429-V as alternate gene silencing products.

For independent verification of ARL8A (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-88429A, sc-88429B and sc-88429C.

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

ARL8A siRNA (h) is recommended for the inhibition of ARL8A 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

ARL8A/B (H-8): sc-398635 is recommended as a control antibody for monitoring of ARL8A 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 ARL8A gene expression knockdown using RT-PCR Primer: ARL8A (h)-PR: sc-88429-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.