

# ARF1 siRNA (h): sc-105086

## BACKGROUND

ADP-ribosylation factors (ARFs), are small guanine nucleotide-binding proteins that enhance the enzymatic activities of cholera toxin, and constitute one family of the RAS superfamily. ARFs are essential and ubiquitous in eukaryotes, as they are involved in vesicular transport and functioning via phospholipase D activation. ARF proteins play a role in membrane traffic and organelle integrity and are intimately tied to their reversible association with membranes and distinct interactions with membrane phospholipids. ARF1 is regulated by the binding and hydrolysis of GTP. Coatamer, or COPI, is a heptameric protein recruited to membranes by ARF1. Research demonstrates that guanine nucleotide exchange-activated ARF1, when located at the Golgi membrane, recruits and binds cytoplasmic COPI to the membranes.

## REFERENCES

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4. Amor, J.C., et al. 1995. Structure of the human ADP-ribosylation factor 1 complexed with GDP. *Nature* 372: 704-708.
5. Hirai, M., et al. 1997. Assignment of human ADP ribosylation factor (ARF) genes ARF1 and ARF3 to chromosomes 1q42 and 12q13, respectively. *Genomics* 34: 263-265.
6. Mossessova, E., et al. 1998. Structure of the guanine nucleotide exchange factor Sec7 domain of human ARNO and analysis of the interaction with ARF GTPase. *Cell* 92: 415-423.
7. Presley, J.F., et al. 2002. Dissection of COPI and ARF1 dynamics *in vivo* and role in Golgi membrane transport. *Nature* 417: 187-193.
8. Renault, L., et al. 2003. Structural snapshots of the mechanism and inhibition of a guanine nucleotide exchange factor. *Nature* 426: 525-530.

## CHROMOSOMAL LOCATION

Genetic locus: ARF1 (human) mapping to 1q42.13.

## PRODUCT

ARF1 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ARF1 shRNA Plasmid (h): sc-105086-SH and ARF1 shRNA (h) Lentiviral Particles: sc-105086-V as alternate gene silencing products.

For independent verification of ARF1 (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-105086A, sc-105086B and sc-105086C.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

ARF1 siRNA (h) is recommended for the inhibition of ARF1 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  $\mu$ M in 66  $\mu$ 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

ARF1 (ARFS 1A9/5): sc-53168 is recommended as a control antibody for monitoring of ARF1 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ARF1 gene expression knockdown using RT-PCR Primer: ARF1 (h)-PR: sc-105086-PR (20  $\mu$ l, 540 bp). 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](http://www.scbt.com) for detailed protocols and support products.