

ARFRP1 siRNA (m): sc-141192

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 and they play a central role in the maintenance of organelle integrity, assembly of coat proteins and activation of phospholipase D (PC-PLD). ARFRP1 (ADP-ribosylation factor related protein 1), also known as ARP or ARL18, is a 201 amino acid membrane-associated GTPase that localizes to the plasma membrane and the Golgi apparatus and is related to the ARF family of regulatory proteins. Expressed in a variety of tissues, ARFRP1 interacts with SYS1 and is thought to be involved in plasma membrane-related signaling events. ARFRP1 exists as multiple alternatively spliced isoforms and is encoded by a gene which maps to a gene cluster on chromosome 20 that is commonly overexpressed in tumors, suggesting a role for ARFRP1 in carcinogenesis.

REFERENCES

- Schürmann, A., et al. 1995. ARP is a plasma membrane-associated Ras-related GTPase with remote similarity to the family of ADP-ribosylation factors. *J. Biol. Chem.* 270: 30657-30663.
- Schürmann, A., et al. 1999. The ADP-ribosylation factor (ARF)-related GTPase ARF-related protein binds to the ARF-specific guanine nucleotide exchange factor cytohesin and inhibits the ARF-dependent activation of phospholipase D. *J. Biol. Chem.* 274: 9744-9751.
- Bai, C., et al. 2000. Overexpression of M68/DcR3 in human gastrointestinal tract tumors independent of gene amplification and its location in a four-gene cluster. *Proc. Natl. Acad. Sci. USA* 97: 1230-1235.
- Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 604699. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
- Behnia, R., et al. 2004. Targeting of the Arf-like GTPase Arl3p to the Golgi requires N-terminal acetylation and the membrane protein Sys1p. *Nat. Cell Biol.* 6: 405-413.
- Shin, H.W., et al. 2005. Roles of ARFRP1 (ADP-ribosylation factor-related protein 1) in post-Golgi membrane trafficking. *J. Cell Sci.* 118: 4039-4048.

CHROMOSOMAL LOCATION

Genetic locus: *Arfrp1* (mouse) mapping to 2 H4.

PRODUCT

ARFRP1 siRNA (m) 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 ARFRP1 shRNA Plasmid (m): sc-141192-SH and ARFRP1 shRNA (m) Lentiviral Particles: sc-141192-V as alternate gene silencing products.

For independent verification of ARFRP1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-141192A, sc-141192B and sc-141192C.

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

ARFRP1 siRNA (m) is recommended for the inhibition of ARFRP1 expression in mouse 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

ARFRP1 (A-8): sc-398568 is recommended as a control antibody for monitoring of ARFRP1 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 ARFRP1 gene expression knockdown using RT-PCR Primer: ARFRP1 (m)-PR: sc-141192-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.