

ARH3 shRNA (h) Lentiviral Particles: sc-78611-V

BACKGROUND

ARH3 (ADP-ribosylhydrolase 3), also known as ADPRHL2 (ADP-ribosylhydrolase like 2), is a 363 amino acid protein that localizes to mitochondria, as well as to both the cytoplasm and the nucleus, and belongs to the ADP-ribosylglycohydrolase family. Expressed ubiquitously, ARH3 uses magnesium as a cofactor to catalyze the hydrolysis of poly(ADP-ribose) that is synthesized after DNA damage. Via its catalytic activity, ARH3 generates ADP-ribose from poly(ADP-ribose) and is thought to play an important role in the maintenance of normal neuronal cell function. The gene encoding ARH3 maps to human chromosome 1, which spans 260 million base pairs, contains over 3,000 genes and comprises nearly 8% of the human genome. Chromosome 1 houses a large number of disease-associated genes, including those that are involved in familial adenomatous polyposis, Stickler syndrome, Parkinson's disease, Gaucher disease, schizophrenia and Usher syndrome. Aberrations in chromosome 1 are found in a variety of cancers, including head and neck cancer, malignant melanoma and multiple myeloma.

REFERENCES

1. Glowacki, G., et al. 2002. The family of toxin-related ecto-ADP-ribosyltransferases in humans and the mouse. *Protein Sci.* 11: 1657-1670.
2. Kernstock, S., et al. 2006. Cloning, expression, purification, crystallization and preliminary X-ray diffraction analysis of human ARH3, the first eukaryotic protein-ADP-ribosylhydrolase. *Acta Crystallogr. Sect. F Struct. Biol. Cryst. Commun.* 62: 224-227.
3. Mueller-Dieckmann, C., et al. 2006. The structure of human ADP-ribosylhydrolase 3 (ARH3) provides insights into the reversibility of protein ADP-ribosylation. *Proc. Natl. Acad. Sci. USA* 103: 15026-15031.

CHROMOSOMAL LOCATION

Genetic locus: ADPRHL2 (human) mapping to 1p34.3.

PRODUCT

ARH3 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see ARH3 siRNA (h): sc-78611 and ARH3 shRNA Plasmid (h): sc-78611-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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

APPLICATIONS

ARH3 shRNA (h) Lentiviral Particles is recommended for the inhibition of ARH3 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

ARH3 (A-7): sc-374162 is recommended as a control antibody for monitoring of ARH3 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 ARH3 gene expression knockdown using RT-PCR Primer: ARH3 (h)-PR: sc-78611-PR (20 μ l, 590 bp). Annealing temperature for the primers should be $55-60^{\circ}$ C and the extension temperature should be $68-72^{\circ}$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.