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Aladin shRNA (h) Lentiviral Particles: sc-45244-V

BACKGROUND

Aladin (Adracalin) belongs to a family of WD repeat-containing proteins. These proteins have a wide variety of functions, including signal transduction regulation, RNA processing and transcription. Aladin plays a role in peripheral and central nervous system development. It is widely expressed, with the highest expression seen in pituitary gland, corpus callosum, cerebellum, adrenal gland and gastrointestinal structures. Defects in Aladin cause the autosomal recessive disorder achalasia-addisonianism-alacrima (triple A) syndrome. Triple A syndrome is characterized by achalasia, alacrima and adrenocorticotropic-resistant adrenal insufficiency. Robust expression in neural systems associated with cognitive, motor and sensory functions is consistent with the myriad of symptoms experienced by patients with triple A syndrome.

REFERENCES

1. Tullio-Pelet, A., et al. 2000. Mutant WD-repeat protein in triple A syndrome. *Nat. Genet.* 26: 332-335.
2. Katsumata, N., et al. 2002. Analysis of the AAAS gene in a Japanese patient with triple A syndrome. *Endocr. J.* 49: 49-53.
3. Houlden, H., et al. 2002. Clinical and genetic characterization of families with triple A (Allgrove) syndrome. *Brain* 125: 2681-2690.
4. Cronshaw, J.M., et al. 2003. The nuclear pore complex protein Aladin is mis-localized in triple A syndrome. *Proc. Natl. Acad. Sci. USA* 100: 5823-5827.
5. Salehi, M., et al. 2005. The diagnosis of adrenal insufficiency in a patient with Allgrove syndrome and a novel mutation in the Aladin gene. *Metabolism* 54: 200-205.
6. Storr, H.L., et al. 2005. Identification of the sites of expression of triple A syndrome mRNA in the rat using *in situ* hybridisation. *Neuroscience* 131: 113-123.

CHROMOSOMAL LOCATION

Genetic locus: AAAS (human) mapping to 12q13.13.

PRODUCT

Aladin 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 Aladin siRNA (h): sc-45244 and Aladin shRNA Plasmid (h): sc-45244-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 or our catalog for detailed protocols and support products.

APPLICATIONS

Aladin shRNA (h) Lentiviral Particles is recommended for the inhibition of Aladin 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

Aladin (B-11): sc-374073 is recommended as a control antibody for monitoring of Aladin 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Aladin gene expression knockdown using RT-PCR Primer: Aladin (h)-PR: sc-45244-PR (20 μ l). 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.