

AHNAK siRNA (m): sc-140916

BACKGROUND

AHNAK (neuroblast differentiation-associated protein AHNAK, desmoyokin) is a 5,890 amino acid protein encoded by the human gene AHNAK. The intronless AHNAK gene is located on human chromosome 11q12 and has three main structural regions: the 251 amino acid N-terminus, a large central region of 4,390 amino acids with multiple repeated units of about 128 amino acids in length, and the 1,002 amino acid C-terminus. The central region seems to have antiparallel β -strands connected by intervening loops. Several putative regulatory elements are clustered within the C-terminal region, including nuclear export localization signals, a leucine zipper, and potential phosphorylation sites for Akt1 and PKC. AHNAK is believed to be an important signalling molecule involved in a wide range of physiological activities and may be required for neuronal cell differentiation. AHNAK also appears to influence β -adrenergic regulation of cardiac L-type Ca^{2+} channel function.

REFERENCES

1. von Boxberg, Y., et al. 2006. Spinal cord injury-induced upregulation of AHNAK, expressed in cells delineating cystic cavities, and associated with neoangiogenesis. *Eur. J. Neurosci.* 24: 1031-1041.
2. De Seranno, S., et al. 2006. Identification of an AHNAK binding motif specific for the Annexin2/S100A10 tetramer. *J. Biol. Chem.* 281: 35030-35038.
3. Haase, H. 2006. AHNAK, a new player in β -adrenergic regulation of the cardiac L-type Ca^{2+} channel. *Cardiovasc. Res.* 73: 19-25.
4. Huang, Y., et al. 2007. AHNAK, a novel component of the dysferlin protein complex, redistributes to the cytoplasm with dysferlin during skeletal muscle regeneration. *FASEB J.* 21: 732-742.

CHROMOSOMAL LOCATION

Genetic locus: Ahnak (mouse) mapping to 19 A.

PRODUCT

AHNAK siRNA (m) is a pool of 2 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see AHNAK shRNA Plasmid (m): sc-140916-SH and AHNAK shRNA (m) Lentiviral Particles: sc-140916-V as alternate gene silencing products.

For independent verification of AHNAK (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-140916A and sc-140916B.

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 μl of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μl of RNase-free water makes a 10 μM solution in a 10 μM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

AHNAK siRNA (m) is recommended for the inhibition of AHNAK 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 μM in 66 μl . 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

AHNAK (1G11): sc-134252 is recommended as a control antibody for monitoring of AHNAK 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 AHNAK gene expression knockdown using RT-PCR Primer: AHNAK (m)-PR: sc-140916-PR (20 μl , 491 bp). Annealing temperature for the primers should be $55-60^{\circ}\text{C}$ and the extension temperature should be $68-72^{\circ}\text{C}$.