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SGK3 shRNA (h) Lentiviral Particles: sc-44852-V

BACKGROUND

Serine/threonine-protein kinase Sgk3 (SGK3), also designated serum/glucocorticoid regulated kinase 3, belongs to the Ser/Thr protein kinase family of proteins. The serum- and glucocorticoid-regulated kinase proteins are closely related to the Akt protein family. SGK1, a homolog of SGK3, activates ion channels, in particular potassium (K⁺) channels. SGK2 and SGK3 have been found to also be involved in this activation process, making all three of these proteins important regulators for cell proliferation, epithelial transport and neuromuscular excitability. SGK3 acts as a mediator of IL-3 dependent survival signals in the cell. It localizes to the early endosome and in vesicle-like structures. SGK3 is a widely expressed protein, but it is primarily detected in kidney, liver, pancreas, brain and heart. Phosphorylation of SGK3 at residue Ser 486 leads to an increase in SGK3 activation.

REFERENCES

1. Dai, F., et al. 1999. Cloning and mapping of a novel human serum/glucocorticoid regulated kinase-like gene, SGKL, to chromosome 8q12.3-q13.1. *Genomics* 62: 95-97.
2. Kobayashi, T., et al. 1999. Characterization of the structure and regulation of two novel isoforms of serum- and glucocorticoid-induced protein kinase. *Biochem. J.* 344: 189-197.
3. Gamper, N., et al. 2002. K⁺ channel activation by all three isoforms of serum- and glucocorticoid-dependent protein kinase SGK. *Pflugers. Arch.* 445: 60-66.
4. Lang, F., et al. 2003. Regulation of channels by the serum and glucocorticoid-inducible kinase-implications for transport, excitability and cell proliferation. *Cell. Physiol. Biochem.* 13: 41-50.
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6. Henke, G., et al. 2004. Regulation of the voltage gated K⁺ channel Kv1.3 by the ubiquitin ligase Nedd4-2 and the serum and glucocorticoid inducible kinase SGK1. *J. Cell. Physiol.* 199: 194-199.

CHROMOSOMAL LOCATION

Genetic locus: SGK3 (human) mapping to 8q13.1.

PRODUCT

SGK3 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 SGK3 siRNA (h): sc-44852 and SGK3 shRNA Plasmid (h): sc-44852-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.

APPLICATIONS

SGK3 shRNA (h) Lentiviral Particles is recommended for the inhibition of SGK3 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

SGK3 (C-6): sc-166847 is recommended as a control antibody for monitoring of SGK3 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-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor SGK3 gene expression knockdown using RT-PCR Primer: SGK3 (h)-PR: sc-44852-PR (20 μ l, 564 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° 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.

PROTOCOLS

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