SANTA CRUZ BIOTECHNOLOGY, INC.

SGK siRNA (X. laevis): sc-77322

BACKGROUND

Serum- and glucocorticoid-regulated kinase (SGK) is a serine/threonine protein kinase and a member of the "AGC" subfamily, which includes protein kinases A, G and C. SGK plays an important role in activating certain potassium, sodium and chloride channels, suggesting an involvement in the regulation of processes such as cell survival, neuronal excitability and renal sodium excretion. SGK contains a catalytic domain, which is most similar to Akt1 (also known as protein kinase B or PKB). SGK is a downstream target of PI 3-kinase-stimulated growth factor signaling, with 3-phosphoinositide-dependent protein kinase 1 (PDK1) capable of phosphorylating the activation-loop of SGK at Threonine 256. The adrenal corticosteroid hormone, Aldosterone, induces the transcription of SGK, which mediates Na^+ transport by stimulating epithelial sodium channel activity. The SGK promoter contains a glucocorticoid response element and an SP-1 regulatory element, and is a transcriptional target for p53. SGK is also a component of the p38 MAPK-mediated response to hyperosmotic stress. The human SGK gene maps to chromosome 6q23.2 and encodes the 431-amino acid SGK protein.

REFERENCES

1. Kleyman, T.R., et al. 1992. Aldosterone does not alter apical cell-surface expression of epithelial Na^+ channels in the amphibian cell line A6. J. Biol. Chem. 267: 9622-9628.
2. Maiyar, A.C., et al. 1996. p53 stimulates promoter activity of the SGK serum/glucocorticoid-inducible serine/threonine protein kinase gene in rodent mammary epithelial cells. J. Biol. Chem. 271: 12414-12422.
3. Waldegger, S., et al. 1997. Cloning and characterization of a putative human serine/threonine protein kinase transcriptionally modified during anisotonic and isotonic alterations of cell volume. Proc. Natl. Acad. Sci. USA 94: 4440-4445.
4. Chen, S.Y., et al. 1999. Epithelial sodium channel regulated by aldosterone-induced protein SGK. Proc. Natl. Acad. Sci. USA 96: 2514-2519.
5. Park, J., et al. 1999. Serum and glucocorticoid-inducible kinase (SGK) is a target of the PI 3-kinase-stimulated signaling pathway. EMBO J. 18: 3024-3033.

CHROMOSOMAL LOCATION

Genetic locus: SGK (X. laevis) mapping to 5L.

PRODUCT

SGK siRNA (X. laevis) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see SGK shRNA Plasmid (X. laevis): sc-77322-SH and SGK shRNA (X. laevis) Lentiviral Particles: sc-77322-V as alternate gene silencing products.

For independent verification of SGK (X. laevis) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-77322A, sc-77322B and sc-77322C.

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

SGK siRNA (X. laevis) is recommended for the inhibition of SGK expression in *Xenopus laevis* 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

SGK (G-4): sc-377360 is recommended as a control antibody for monitoring of SGK 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 SGK gene expression knockdown using RT-PCR Primer: SGK (X. laevis)-PR: sc-77322-PR (20 µl, 449 bp). 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.