SANTA CRUZ BIOTECHNOLOGY, INC.

KANK1 siRNA (h): sc-92843

BACKGROUND

KANK1 (KN motif and ankyrin repeat domains 1), also known as ANKRD15, is a 1,352 amino acid protein containing five ANK domains. KANK1 interacts with 14-3-3, regulated by Insulin and EGF and mediated through phosphorylation of Kank by Akt, which inhibits insultin-induced cell migration as well as Insulin and active Akt-dependent activation of RhoA. KANK1 also negatively regulates the formation of actin stress fibbers through inhibition of RhoA activity. KANK1 also interacts with IRSp53, inhibiting the binding of IRSp53 with active Rac1 which in turn inhibits the development of lamellipodia but not filopodia. KANK1 also regulates cell polarity during directed migration in wound healing. KANK1 is also thought to inhibit fibronectin-mediated cell spreading and neurite outgrowth. Mutations in the KANK1 gene results in CPSQ2 (cerebral palsy, spastic quadriplegic 2), a non-progressive disorder of movement and/or posture resulting from defects in the developing nervous system.

REFERENCES

1. Wang, Y., et al. 2005. Alternative splicing of the human Kank gene produces two types of Kank protein. Biochem. Biophys. Res. Commun. 330: 1247-1253.
2. Roy, B.C., et al. 2005. Pathological characterization of Kank in renal cell carcinoma. Exp. Mol. Pathol. 78: 41-48.
3. Wang, Y., et al. 2006. Nucleo-cytoplasmic shuttling of human Kank protein accompanies intracellular translocation of β-catenin. J. Cell Sci. 119: 4002-4010.
4. Kakinuma, N., et al. 2008. Kank regulates RhoA-dependent formation of Actin stress fibers and cell migration via 14-3-3 in PI3K-Akt signaling. J. Cell Biol. 181: 537-549.
5. Roy, B.C., et al. 2009. Kank attenuates Actin remodeling by preventing interaction between IRSp53 and Rac1. J. Cell Biol. 184: 253-267.
6. Medves, S., et al. 2010. KANK1, a candidate tumor suppressor gene, is fused to PDGFRB in an imatinib-responsive myeloid neoplasm with severe thrombocythemia. Leukemia 24: 1052-1055.

CHROMOSOMAL LOCATION

Genetic locus: KANK1 (human) mapping to 9p24.3.

PRODUCT

KANK1 siRNA (h) 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 KANK1 shRNA Plasmid (h): sc-92843-SH and KANK1 shRNA (h) Lentiviral Particles: sc-92843-V as alternate gene silencing products.

For independent verification of KANK1 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-92843A, sc-92843B and sc-92843C.

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

KANK1 siRNA (h) is recommended for the inhibition of KANK1 expression in human 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

KANK1 (177D2T): sc-517629 is recommended as a control antibody for monitoring of KANK1 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 KANK1 gene expression knockdown using RT-PCR Primer: KANK1 (h)-PR: sc-92843-PR (20 µl). 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.