SANTA CRUZ BIOTECHNOLOGY, INC.

NCKX1 siRNA (h): sc-61158

BACKGROUND

NCKX1, also designated solute carrier family 24, member 1 (SLC24A1) or sodium/calcium/potassium exchanger 1, belongs to a family of potassium-dependent sodium/calcium exchangers. Members of this group of proteins contain two large hydrophilic loops and two sets of multiple transmembrane-spanning segments. One intron in the 5' untranslated region and eight within the coding region of the NCKX1 gene have been identified; exon length varies from 54 to 2,037 bp. Human NCKX1 encodes a protein of 1,081 amino acids that shows 64% overall identity with the cow protein. The two sets of presumed transmembrane domains and their short connecting loops show 94% identity with that of the bovine, while the extracellular loop at the amino terminus is only 59% identical.

REFERENCES

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2. Poon, S., Leach, S., Li, X.F., Tucker, J.E., Schnetkamp, P.P. and Lytton, J. 2000. Alternatively spliced isoforms of the rat eye sodium/calcium + potassium exchanger NCKX1. Am. J. Physiol., Cell Physiol. 278: C651-C660.
3. Kang, K. and Schnetkamp, P.P. 2003. Signal sequence cleavage and plasma membrane targeting of the retinal rod NCKX1 and cone NCKX2 Na^{+}/Ca^{2+}-K^{+} exchangers. Biochemistry 42: 9438-9445.
4. Aneiros, E., Philipp, S., Lis, A., Freichel, M. and Cavalie, A. 2005. Modulation of Ca^{2+} signaling by Na^{+}/Ca^{2+} exchangers in mast cells. J. Immunol. 174: 119-130.
5. Kang, K.J., Shibukawa, Y., Szerencsei, R.T. and Schnetkamp, P.P. 2005. Substitution of a single residue, Asp 575, renders the NCKX2 K^{+}-dependent Na^{+}/Ca^{2+} exchanger independent of K^{+}. J. Biol. Chem. 280: 6834-6839.

CHROMOSOMAL LOCATION

Genetic locus: SLC24A1 (human) mapping to 15q22.31.

PRODUCT

NCKX1 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 NCKX1 shRNA Plasmid (h): sc-61158-SH and NCKX1 shRNA (h) Lentiviral Particles: sc-61158-V as alternate gene silencing products.

For independent verification of NCKX1 (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-61158A, sc-61158B and sc-61158C.

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

NCKX1 siRNA (h) is recommended for the inhibition of NCKX1 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

NCKX1 (E-6): sc-393400 is recommended as a control antibody for monitoring of NCKX1 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 NCKX1 gene expression knockdown using RT-PCR Primer: NCKX1 (h)-PR: sc-61158-PR (20 µl, 566 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.