SANTA CRUZ BIOTECHNOLOGY, INC.

ELKS siRNA (h): sc-60572

BACKGROUND

The glutamine, leucine, lysine, and serine-rich protein ELKS, also designated Rab6-interacting protein 2 and CAZ-associated structural protein (CAST) or the acronym ERC, is a member of a family of RIM-binding proteins. RIMs are presynaptic active zone proteins that regulate neurotransmitter release. This class of protein functions by recruiting IκBα to the IκB kinase (IKK) complex, and thus serves a regulatory function for IKK activation. Five isoforms of ELKS (α, β, γ, δ, and ε) exist and have multiple coding region differences and distinct C-termini. Only brain-specific ELKS bind to RIMs, but both ubiquitous and brain-specific ELKS bind to Rab6, a GTP-binding protein involved in membrane traffic at the Golgi complex. Fusion of ELKS to RET due to translocation t(10;12)(q11;p13) results in a papillary thyroid carcinoma.

REFERENCES

1. Nakata, T., et al. 1999. Fusion of a novel gene, ELKS, to RET due to translocation t(10;12)(q11;p13) in a papillary thyroid carcinoma. Genes Chromosomes Cancer 25: 97-103.
2. Wang, Y., et al. 2002. A family of RIM-binding proteins regulated by alternative splicing: implications for the genesis of synaptic active zones. Proc. Natl. Acad. Sci. USA 99: 14464-14469.
3. Ko, J., et al. 2003. Interaction of the ERC family of multidomain proteins. J. Biol. Chem. 278: 42377-42385.
4. Deguchi-Tawarada, M., et al. 2004. CAST2: identification and characterization of a protein structurally related to the presynaptic cytomatrix protein CAST. Genes Cells 9: 15-23.
5. Deken, S.L., et al. 2005. Redundant localization mechanisms of RIM and ELKS in *Caenorhabditis elegans*. J. Neurosci. 25: 5975-5983.
6. Ohara-Imaizumi, M., et al. 2005. ELKS, a protein structurally related to the active zone-associated protein CAST, is expressed in pancreatic β cells and functions in Insulin exocytosis: interaction of ELKS with exocytotic machinery analyzed by total internal reflection fluorescence microscopy. Mol. Biol. Cell 16: 3289-3300.
7. Lu, J., et al. 2005. Solution structure of the RIM1α PDZ domain in complex with an ELKS1b C-terminal peptide. J. Mol. Biol. 352: 455-466.

CHROMOSOMAL LOCATION

Genetic locus: ERC1 (human) mapping to 12p13.33.

PRODUCT

ELKS 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 ELKS shRNA Plasmid (h): sc-60572-SH and ELKS shRNA (h) Lentiviral Particles: sc-60572-V as alternate gene silencing products.

For independent verification of ELKS (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-60572A, sc-60572B and sc-60572C.

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

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

ELKS (G-10): sc-515041 is recommended as a control antibody for monitoring of ELKS 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 ELKS gene expression knockdown using RT-PCR Primer: ELKS (h)-PR: sc-60572-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.