SANTA CRUZ BIOTECHNOLOGY, INC.

INSC siRNA (h): sc-96461

BACKGROUND

INSC (protein inscuteable homolog) is a 579 amino acid protein that interacts with PAR-3, AGS3, nucleobindin and PARD3B. INSC may function as an adapter linking the Par3 complex to the AGS3/nucleobindin complex. Involved in spindle orientation during mitosis, INSC may also regulate differentiation and cell proliferation in the developing nervous system, play a role in the asymmetric division of fibroblasts and participate in the process of stratification of the squamous epithelium. Localized to the cytoplasm, INSC is expressed in fetal cochlea and exists as five isoforms due to alternative splicing events. Isoform 1 is expressed in various tissues, with stronger expression in liver, kidney and small intestine, while isoform 2 is abundantly expressed in small intestine, with lower levels in lung and pancreas.

REFERENCES

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2. Katoh, M., et al. 2003. Identification and characterization of human Inscuteable gene in silico. Int. J. Mol. Med. 11: 111-116.
3. Zigman, M., et al. 2005. Mammalian inscuteable regulates spindle orientation and cell fate in the developing retina. Neuron 48: 539-545.
4. Izaki, T., et al. 2006. Two forms of human Inscuteable-related protein that links Par3 to the Pins homologues LGN and AGS3. Biochem. Biophys. Res. Commun. 341: 1001-1006.
5. Online Mendelian Inheritance in Man, OMIM™. 2006. Johns Hopkins University, Baltimore, MD. MIM Number: 610668. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
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CHROMOSOMAL LOCATION

Genetic locus: INSC (human) mapping to 11p15.2.

PRODUCT

INSC 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 INSC shRNA Plasmid (h): sc-96461-SH and INSC shRNA (h) Lentiviral Particles: sc-96461-V as alternate gene silencing products.

For independent verification of INSC (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-96461A, sc-96461B and sc-96461C.

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

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

INSC (G-6): sc-514398 is recommended as a control antibody for monitoring of INSC 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 IgM-HRP: sc-2064 (dilution range: 1:500-1:5,000), TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgM-FITC: sc-2082 (dilution range: 1:100-1:400) or goat anti-mouse IgM-TR: sc-2983 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor INSC gene expression knockdown using RT-PCR Primer: INSC (h)-PR: sc-96461-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 or our catalog for detailed protocols and support products.