SANTA CRUZ BIOTECHNOLOGY, INC.

N-Shc siRNA (h): sc-40975

BACKGROUND

Src homology (SH2) domains are noncatalytic sequences that are conserved among a number of cytoplasmic signaling proteins. These signaling proteins are directly regulated by receptor tyrosine kinases and control the activation of mitogenic signal transduction pathways by such receptors. For instance, ligand-induced activation of the EGF and PDGF receptors induces dimerization, triggers receptor autophosphorylation on tyrosine residues and results in the binding of a number of cytoplasmic SH2 domain proteins such as PLC γ1, Ras GAP and PI 3-kinase to the activated receptors. Another gene, Shc, encodes two proteins with a single SH2 domain. A Shc-related gene N-Shc (for neuronal Shc), encodes a protein that contains two phosphotyrosine domains (PTB), a single SH2 domain and is expressed exclusively in the brain. Neither Shc nor N-Shc have any identifiable catalytic activity, suggesting them to be members of an expanding class of proteins that function to couple activated growth factor receptors to downstream signaling intermediates.

REFERENCES

1. Ullrich, A., et al. 1990. Signal transduction by receptors with tyrosine kinase activity. Cell 61: 203-212.
2. Morrison, D.K., et al. 1990. Platelet-derived growth factor (PDGF)-dependent association of phospholypase C-γ with the PDGF receptor signaling complex. Mol. Cell. Biol. 10: 2359-2366.
3. Cantley, L.C., et al. 1991. Oncogenes and signal transduction. Cell 64: 281-302.
4. Koch, C.A., et al. 1991. SH2 and SH3 domains: elements that control interactions of cytoplasmic signaling proteins. Science 252: 669-674.
5. McGlade, J., et al. 1992. Shc proteins are phosphorylated and regulated by the v-Src and v-Fps protein-tyrosine kinases. Proc. Natl. Acad. Sci. USA 89: 8869-8873.
6. Pelicci, G., et al. 1992. A novel transforming protein (SHC) with an SH2 domain is implicated in mitogenic signal transduction. Cell 70: 93-104.
7. Ravichandran, K.S., et al. 1993. Interaction of Shc with the ζ chain of the T cell receptor upon T cell activation. Science 262: 902-905.

CHROMOSOMAL LOCATION

Genetic locus: SHC3 (human) mapping to 9q22.1.

PRODUCT

N-Shc 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 N-Shc shRNA Plasmid (h): sc-40975-SH and N-Shc shRNA (h) Lentiviral Particles: sc-40975-V as alternate gene silencing products.

For independent verification of N-Shc (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-40975A, sc-40975B and sc-40975C.

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

N-Shc siRNA (h) is recommended for the inhibition of N-Shc 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

N-Shc (H-7): sc-365598 is recommended as a control antibody for monitoring of N-Shc 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 N-Shc gene expression knockdown using RT-PCR Primer: N-Shc (h)-PR: sc-40975-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.