SANTA CRUZ BIOTECHNOLOGY, INC.

Nischarin siRNA (m): sc-61202

BACKGROUND

Integrins play important roles in key cellular functions, including cytoskeletal organization, growth, survival, motility and gene expression regulation. Nischarin is a novel intracellular protein, that binds to the cytoplasmic domain of integrin $\alpha 5/\beta 1$ and interacts with various members of the PAK family of kinases. Nischarin binding to PAK1 inhibits the ability of PAK1 to phosphorylate substrates. When bound, this complex localizes to membrane ruffles which are involved in cell motility. Nischarin also acts as an antagonist of Rac function on cell movement and alters actin filament organization. These functions give Nischarin a possible role in cell migration regulation. Nischarin is a primarily cytoplasmic protein primarily expressed in kidney and brain.

REFERENCES

1. Dontenwill, M., et al. 2003. IRAS is an anti-apoptotic protein. Ann. N.Y. Acad. Sci. 1009: 400-412.
2. Chen, M.J., et al. 2003. Intracellular effect of imidazoline receptor on α_{2A}-noradrenergic receptor. Ann. N.Y. Acad. Sci. 1009: 427-438.
3. Zhu, H., et al. 2003. Relationship between platele imidazoline-1 receptor, IRAS. Ann. N.Y. Acad. Sci. 1009: 439-446.
4. Lim, K.P., et al. 2004. Human Nischarin/imidazoline receptor antisera-selected protein is targeted to the endosomes by a combined action of a PX domain and a coiled-coil region. J. Biol. Chem. 279: 54770-54782.
5. Alahari, S.K., et al. 2004. The integrin-binding protein Nischarin regulates cell migration by inhibiting PAK. EMBO J. 23: 2777-2788.

CHROMOSOMAL LOCATION

Genetic locus: Nisch (mouse) mapping to 14 B.

PRODUCT

Nischarin siRNA (m) 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 Nischarin shRNA Plasmid (m): sc-61202-SH and Nischarin shRNA (m) Lentiviral Particles: sc-61202-V as alternate gene silencing products.

For independent verification of Nischarin (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-61202A, sc-61202B and sc-61202C.

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

Nischarin siRNA (m) is recommended for the inhibition of Nischarin expression in mouse 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

Nischarin (F-3): sc-374408 is recommended as a control antibody for monitoring of Nischarin 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 Nischarin gene expression knockdown using RT-PCR Primer: Nischarin (m)-PR: sc-61202-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.