

Neurogranin siRNA (m): sc-42075

BACKGROUND

Neurogranin (formerly designated p17, also known as RC3 and BICKS) is a neuron-specific substrate for protein kinase C (PKC). Neurogranin is a post-synaptic protein that is highly enriched in brain, with restricted expression in the cortex, striatum, hippocampus, thalamus, hypothalamus and olfactory bulb nuclei. Neurogranin binds calmodulin at low levels of calcium, thereby regulating calmodulin-dependent nitric oxide synthase. Conversely, nitric oxide modifies Neurogranin, reducing its ability to bind calmodulin or to be phosphorylated by PKC. This phosphorylation site domain is adjacent to the predicted calmodulin-binding region.

REFERENCES

1. Baudier, J., et al. 1989. Protein kinase C substrates from bovine brain. Purification and characterization of Neuromodulin, a neuron-specific calmodulin-binding protein. *J. Biol. Chem.* 264: 1824-1828.
2. Watson, J.B., et al. 1990. Subtractive cDNA cloning of RC3, a rodent cortex-enriched mRNA encoding a novel 78 residue protein. *J. Neurosci. Res.* 26: 397-408.
3. Baudier, J., et al. 1991. Purification and characterization of a brain-specific protein kinase C substrate, Neurogranin. Identification of a consensus amino acid sequence between Neurogranin and Neuromodulin (GAP-43) that corresponds to the protein kinase C phosphorylation site and the calmodulin-binding domain. *J. Biol. Chem.* 266: 229-237.
4. Watson, J.B., et al. 1992. Localization of the protein kinase C phosphorylation/calmodulin-binding substrate RC3 in dendritic spines of neostriatal neurons. *Proc. Natl. Acad. Sci. USA* 89: 8581-8585.
5. Huang, K.P., et al. 1993. Characterization of a 7.5-kDa protein kinase C substrate (RC3 protein, Neurogranin) from rat brain. *Arch. Biochem. Biophys.* 305: 570-580.
6. Martzen, M.R., et al. 1995. The dendritic peptide Neurogranin can regulate a calmodulin-dependent target. *J. Neurochem.* 64: 92-100.
7. Sheu, F.S., et al. 1996. Nitric oxide modification of rat brain Neurogranin affects its phosphorylation by protein kinase C and affinity for calmodulin. *J. Biol. Chem.* 271: 22407-22413.

CHROMOSOMAL LOCATION

Genetic locus: Nrgn (mouse) mapping to 9 A4.

PRODUCT

Neurogranin 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Neurogranin shRNA Plasmid (m): sc-42075-SH and Neurogranin shRNA (m) Lentiviral Particles: sc-42075-V as alternate gene silencing products.

For independent verification of Neurogranin (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-42075A, sc-42075B and sc-42075C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCL, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Neurogranin siRNA (m) is recommended for the inhibition of Neurogranin 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 μ M in 66 μ l. 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

Neurogranin (C-7): sc-515092 is recommended as a control antibody for monitoring of Neurogranin 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 Neurogranin gene expression knockdown using RT-PCR Primer: Neurogranin (m)-PR: sc-42075-PR (20 μ l). 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.