SANTA CRUZ BIOTECHNOLOGY, INC.

RIC-3 siRNA (h): sc-72301

BACKGROUND

RIC-3 (resistant to inhibitors of cholinesterase-3) is the mammalian homolog of the ric-3 protein from *C. elegans*. It contains two transmembrane domains and a coiled coil domain. RIC-3 is expressed in neurons and localizes to the endoplasmic reticulum where it plays a role in receptor folding and subunit assembly. In particular, RIC-3 is a nicotinic acetylcholine receptor (nAChR)-associated protein and it significantly enhances the subunit assembly, proper folding, stability and surface expression of several heteromeric and homomeric nAChR subtypes as well as some 5-HT3 receptors. This suggests that RIC-3 may be an important regulator of receptor expression. Several isoforms exist for RIC-3 and they exhibit overlapping but distinct localizations. In addition, these isoforms may have various affects on receptor expression.

REFERENCES

1. Halevi, S., et al. 2003. Conservation within the RIC-3 gene family. Effectors of mammalian nicotinic acetylcholine receptor expression. J. Biol. Chem. 278: 34411-34417.
2. Williams, M.E., et al. 2005. RIC-3 promotes functional expression of the nicotinic acetylcholine receptor α7 subunit in mammalian cells. J. Biol. Chem. 280: 1257-1263.
3. Cheng, A., et al. 2005. Cell surface expression of 5-hydroxytryptamine type 3 receptors is promoted by RIC-3. J. Biol. Chem. 280: 22502-22507.
4. Castillo, M., et al. 2005. Dual role of the RIC-3 protein in trafficking of serotonin and nicotinic acetylcholine receptors. J. Biol. Chem. 280: 27062-27068.
5. Ben-Ami, H.C., et al. 2005. RIC-3 affects properties and quantity of nicotinic acetylcholine receptors via a mechanism that does not require the coiled-coil domains. J. Biol. Chem. 280: 28053-28060.
6. Lansdell, S.J., et al. 2005. RIC-3 enhances functional expression of multiple nicotinic acetylcholine receptor subtypes in mammalian cells. Mol. Pharmacol. 68: 1431-1438.
7. Severance, E.G. and Yolken, R.H. 2007. Lack of RIC-3 congruence with β2 subunit-containing nicotinic acetylcholine receptors in bipolar disorder. Neuroscience 148: 454-460.

CHROMOSOMAL LOCATION

Genetic locus: RIC3 (human) mapping to 11p15.4.

PRODUCT

RIC-3 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 RIC-3 shRNA Plasmid (h): sc-72301-SH and RIC-3 shRNA (h) Lentiviral Particles: sc-72301-V as alternate gene silencing products.

For independent verification of RIC-3 (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-72301A, sc-72301B and sc-72301C.

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

RIC-3 siRNA (h) is recommended for the inhibition of RIC-3 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

RIC-3 (G-8): sc-377408 is recommended as a control antibody for monitoring of RIC-3 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 RIC-3 gene expression knockdown using RT-PCR Primer: RIC-3 (h)-PR: sc-72301-PR (20 µl, 572 bp). 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.