SANTA CRUZ BIOTECHNOLOGY, INC.

RGL1 siRNA (h): sc-62936

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

Ral GDS (Ral guanine nucleotide dissociation stimulator) is a guanine nucleotide exchange factor (GEF) that activates Ral and is implicated in oncogenic Ras-induced cell transformation. RGL1 (Ral guanine nucleotide dissociation stimulator-like 1), also known as RGL or RalGDS-like 1, is a 768 amino acid protein that is a putative GEF. Strongly expressed in brain, heart, spleen, kidney and testis, RGL1 is a downstream effector protein that is involved in Ras and Ral signaling pathways. RGL1 contains an N-terminal Ras-GEF domain and a C-terminal Ras-interacting domain that interacts with the GTP-bound form of Ras through its effector loop. Due to its similarity to Ral GDS, RGL1 may be implicated in carcinogenesis. Two isoforms exist due to alternative splicing events.

REFERENCES

1. Wolthuis, R.M., et al. 1996. RalGDS-like factor (Rlf) is a novel Ras and Rap 1A-associating protein. Oncogene 13: 353-362.
2. Murai, H., et al. 1997. Characterization of Ral GDP dissociation stimulator-like (RGL) activities to regulate c-Fos promoter and the GDP/GTP exchange of Ral. J. Biol. Chem. 272: 10483-10490.
3. Shirouzu, M., et al. 1999. Double-mutant analysis of the interaction of Ras with the Ras-binding domain of RGL. Biochemistry 38: 5103-5110.
4. Sood, R., et al. 2000. The human RGL (RalGDS-like) gene: cloning, expression analysis and genomic organization. Biochim. Biophys. Acta 1491: 285-288.
5. Wen, C.K. and Chang, C. 2002. *Arabidopsis* RGL1 encodes a negative regulator of gibberellin responses. Plant Cell 14: 87-100.
6. González-García, A., et al. 2005. Ral GDS is required for tumor formation in a model of skin carcinogenesis. Cancer Cell 7: 219-226.
7. Ryu, C.H., et al. 2005. The merlin tumor suppressor interacts with Ral guanine nucleotide dissociation stimulator and inhibits its activity. Oncogene 24: 5355-5364.
8. Busov, V., et al. 2006. Transgenic modification of gai or RGL1 causes dwarfing and alters gibberellins, root growth, and metabolite profiles in *Populus*. Planta 224: 288-299.

CHROMOSOMAL LOCATION

Genetic locus: RGL1 (human) mapping to 1q25.3.

PRODUCT

RGL1 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 RGL1 shRNA Plasmid (h): sc-62936-SH and RGL1 shRNA (h) Lentiviral Particles: sc-62936-V as alternate gene silencing products.

For independent verification of RGL1 (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-62936A, sc-62936B and sc-62936C.

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

RGL1 siRNA (h) is recommended for the inhibition of RGL1 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

RGL1 (G-2): sc-377170 is recommended as a control antibody for monitoring of RGL1 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 RGL1 gene expression knockdown using RT-PCR Primer: RGL1 (h)-PR: sc-62936-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.