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DANGER siRNA (m): sc-142872

BACKGROUND

The inositol 1,4,5-triphosphate receptor, IP3R, acts as an inositol triphosphate (IP3)-gated calcium release channel in a variety of cell types. Three IP3 receptor subtypes have been described and are designated IP3R-I, IP3R-II and IP3R-III. DANGER, also known as inositol 1,4,5-triphosphate receptor-interacting protein (ITPRIP) or KIAA1754, is a 547 amino acid protein that interacts with IP3 receptors. Localized to the cell membrane, DANGER enhances Ca²⁺-regulated inhibition of IP3R Ca²⁺ release. The gene that encodes DANGER maps to human chromosome 10. Spanning nearly 135 million base pairs, chromosome 10 makes up approximately 4.5% of total DNA in cells and encodes nearly 1,200 genes. Several protein-coding genes, including those that encode for chemokines, cadherins, excision repair proteins, early growth response factors (Egrs) and fibroblast growth receptors (FGFRs), are located on chromosome 10.

REFERENCES

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3. Cameron, A.M., et al. 1995. Calcineurin associated with the inositol 1,4,5-triphosphate receptor-FKBP12 complex modulates Ca²⁺ flux. *Cell* 83: 463-472.
4. Deloukas, P., French, L., Meitinger, T. and Moschonas, N.K. 2000. Report of the third international workshop on human chromosome 10 mapping and sequencing 1999. *Cytogenet. Cell Genet.* 90: 1-12.
5. van Rossum, D.B., Patterson, R.L., Cheung, K.H., Barrow, R.K., Syrovatkin, V., Gessell, G.S., Burkholder, S.G., Watkins, D.N., Foskett, J.K. and Snyder, S.H. 2006. DANGER, a novel regulatory protein of inositol 1,4,5-triphosphate-receptor activity. *J. Biol. Chem.* 281: 37111-37116.

CHROMOSOMAL LOCATION

Genetic locus: Itprp (mouse) mapping to 19 D1.

PRODUCT

DANGER 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 DANGER shRNA Plasmid (m): sc-142872-SH and DANGER shRNA (m) Lentiviral Particles: sc-142872-V as alternate gene silencing products.

For independent verification of DANGER (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-142872A, sc-142872B and sc-142872C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

DANGER siRNA (m) is recommended for the inhibition of DANGER 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

DANGER (H-7): sc-514861 is recommended as a control antibody for monitoring of DANGER 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 DANGER gene expression knockdown using RT-PCR Primer: DANGER (m)-PR: sc-142872-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.