DENND4B siRNA (h): sc-88449



The Power to Question

BACKGROUND

DENND4B (DENN/MADD domain containing 4B), also known as KIAA0476, is a 1,496 amino acid protein that contains a dDENN domain, a DENN domain, a uDENN domain, a MABP domain and two PPR (pentatricopeptide) repeats. The DENND4B protein is thought to target a tubular membrane compartment adjacent to the Golgi, where it activates Rab10. The gene encoding DENND4B maps to human chromosome 1, the largest human chromosome which spans about 260 million base pairs and makes up 8% of the human genome. Other notable genes located on chromosome 1 include LMNA, which is associated with the rare aging disease Hutchinson-Gilford progeria, and the MUTYH gene, which is partially responsible for familial adenomatous polyposis.

REFERENCES

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- Blackwood, D.H., et al. 2001. Schizophrenia and affective disorders-cosegregation with a translocation at chromosome 1q42 that directly disrupts brain-expressed genes: clinical and P300 findings in a family. Am. J. Hum. Genet. 69: 428-433.
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- 6. Lans, H., et al. 2006. Cell biology: aging nucleus gets out of shape. Nature 440: 32-34.
- McClintock, D., et al. 2006. Hutchinson-Gilford progeria mutant lamin A primarily targets human vascular cells as detected by an anti-lamin A G608G antibody. Proc. Natl. Acad. Sci. USA 103: 2154-2159.

CHROMOSOMAL LOCATION

Genetic locus: DENND4B (human) mapping to 1q21.3.

PRODUCT

DENND4B 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see DENND4B shRNA Plasmid (h): sc-88449-SH and DENND4B shRNA (h) Lentiviral Particles: sc-88449-V as alternate gene silencing products.

For independent verification of DENND4B (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-88449A, sc-88449B and sc-88449C.

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

DENND4B siRNA (h) is recommended for the inhibition of DENND4B 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

DENND4B (F-10): sc-514660 is recommended as a control antibody for monitoring of DENND4B 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 DENND4B gene expression knockdown using RT-PCR Primer: DENND4B (h)-PR: sc-88449-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.

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