

fidgetin siRNA (h): sc-62318

BACKGROUND

The mouse Fign gene encodes a 759 amino acid protein that is a member of the AAA (ATPases associated with diverse cellular activities) family of ATPases. Fidgetin is a member of the "meiotic" or subfamily-7 group of ATPases associated with diverse cellular activities (AAA proteins). Fidgetin can interact with itself and this interaction can be abolished by truncating either the N- or C-terminus of the protein. AAA proteins are molecular chaperones that facilitate membrane fusion, proteolysis, peroxisome biogenesis, endosome sorting and meiotic spindle formation. The mouse mutation fidget arose spontaneously in a heterogeneous albino stock. This mutant mouse is characterized by a side-to-side head-shaking and circling behavior due to reduced or absent semicircular canals. Fidget mice have small eyes that are associated with cell-cycle delay and insufficient growth of the retinal neural epithelium, and lower penetrance skeletal abnormalities including pelvic girdle dysgenesis, skull bone fusions and polydactyly.

REFERENCES

1. Cox, G.A., et al. 2000. The mouse fidgetin gene defines a new role for AAA family proteins in mammalian development. *Nat. Genet.* 26: 198-202.
2. Yakushiji, Y., et al. 2004. Identification of a cysteine residue important for the ATPase activity of *C. elegans* fidgetin homologue. *FEBS Lett.* 578: 191-197.
3. Yang, Y., et al. 2005. Functional characterization of fidgetin, an AAA-family protein mutated in fidget mice. *Exp. Cell Res.* 304: 50-58.
4. Yakushiji, Y., et al. 2006. Mutational analysis of the functional motifs in the ATPase domain of *Caenorhabditis elegans* fidgetin homologue FIGL-1: firm evidence for an intersubunit catalysis mechanism of ATP hydrolysis by AAA ATPases. *J. Struct. Biol.* 156: 93-100.
5. Yang, Y., et al. 2006. Interaction between fidgetin and protein kinase A-anchoring protein AKAP95 is critical for palatogenesis in the mouse. *J. Biol. Chem.* 281: 22352-22359.

CHROMOSOMAL LOCATION

Genetic locus: FIGN (human) mapping to 2q24.3.

PRODUCT

fidgetin 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 fidgetin shRNA Plasmid (h): sc-62318-SH and fidgetin shRNA (h) Lentiviral Particles: sc-62318-V as alternate gene silencing products.

For independent verification of fidgetin (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-62318A, sc-62318B and sc-62318C.

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

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

fidgetin (F-2): sc-514956 is recommended as a control antibody for monitoring of fidgetin 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 fidgetin gene expression knockdown using RT-PCR Primer: fidgetin (h)-PR: sc-62318-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.