

Scrib siRNA (m): sc-36467

BACKGROUND

Drosophila melanogaster genes, which are categorized based on the type of protein for which they encode, represent six major classifications, including intracellular signaling proteins, transmembrane proteins, RNA binding proteins, secreted factors, transcription regulators (basic helix-loop-helix, homeo-domain containing, zinc finger containing and chromatin associated) and other functional proteins. Morphogenesis and cell differentiation in *Drosophila* requires accurate control of cell division. Discs large (Dlg), Scribble (Scrib) and Lethal giant larvae (Lgl) tumor suppressor proteins regulate multiple aspects of neuroblast asymmetric cell division. Dlg/Scrib/Lgl proteins show apical cortical enrichment at prophase/metaphase and have a uniform cortical distribution. Mutations in the genes encoding multi-PDZ (PSD-95, Discs-large and ZO-1) and the leucine-rich-repeat protein Scrib cause aberrant cell shapes and the loss of monolayer organization of embryonic epithelia. The human homolog, hScrib, is intracellularly localized to the vertebrate tight junction, which functions to correctly place adherens junctions. The PDZ domains of Scrib are predicted to bind to the consensus S/TXV at the C-terminus of proteins. PDZ domain proteins have been implicated at several different sites of the protein trafficking pathway, suggesting that Scrib is required for the localization of several epithelial determinants.

REFERENCES

1. Lehner, C.F. 1991. Pulling the string: cell cycle regulation during *Drosophila* development. *Semin. Cell Biol.* 2: 223-231.
2. Songyang, Z., et al. 1997. Recognition of unique carboxyl-terminal motifs by distinct PDZ domains. *Science* 275: 73-77.
3. Balda, M.S. and Matter, K. 1998. Tight junctions. *J. Cell Sci.* 111: 541-547.
4. Jou, T.S., et al. 1998. Structural and functional regulation of tight junctions by Rho A and Rac 1 small GTPases. *J. Cell Biol.* 142: 101-115.
5. Adams, M.D., et al. 2000. The genome sequence of *Drosophila melanogaster*. *Science* 287: 2185-2195.
6. Bilder, D. and Perrimon, N. 2000. Localization of apical epithelial determinant by the basolateral PDZ protein Scribble. *Nature* 403: 676-680.
7. Roche, J.P., et al. 2002. Regulation of synaptic plasticity and synaptic vesicle dynamics by the PDZ protein Scribble. *J. Neurosci.* 22: 6471-6479.

CHROMOSOMAL LOCATION

Genetic locus: Scrib (mouse) mapping to 15 D3.

PRODUCT

Scrib 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 Scrib shRNA Plasmid (m): sc-36467-SH and Scrib shRNA (m) Lentiviral Particles: sc-36467-V as alternate gene silencing products.

For independent verification of Scrib (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-36467A, sc-36467B and sc-36467C.

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

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

Scrib (D-2): sc-374139 is recommended as a control antibody for monitoring of Scrib 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 Scrib gene expression knockdown using RT-PCR Primer: Scrib (m)-PR: sc-36467-PR (20 μ l, 443 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.