SANTA CRUZ BIOTECHNOLOGY, INC.

PARD6B siRNA (m): sc-62752

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The Power to Question

BACKGROUND

Cellular asymmetry is critical for the development of multicellular organisms. PARD (partitioning-defective) proteins play important roles in asymmetric cell division and polarized growth, whereas Cdc42 and Rac mediate establishment of cell growth and polarity and contribute to oncogenic transformation by Ras. The human PARD6, a 345 amino acid polypeptide, has a PDZ domain and a CRIB-like (Cdc42/Rac interactive binding) motif. PARD6 interacts with GTP-bound Rac and Cdc42 via this motif and with the atypical PKC isoforms PKCι/λ and PKCζ via N-terminal head to head association. These interactions allow formation of a ternary complex *in vitro* and *in vivo,* which is implicated in the formation of normal tight junctions at epithelial cell-cell contacts and is also involved in the polarization of mother cells before asymmetric cell division in *C. elegans*. PARD6 acts through PARD3 by localizing or maintaining the PARD3 protein at the cell periphery. PARD6A, also designated PAR-6α, PAR6C, TAX40 and TIP-40, is expressed in pancreas, skeletal muscle, brain and heart, and is weakly expressed in kidney and placenta. PAR6B is expressed in pancreas and in both adult and fetal kidney, and is weakly expressed in placenta and lung.

REFERENCES

1. Watts, J.L., et al. 1996. PAR-6, a gene involved in the establishment of asymmetry in early *C. elegans* embryos, mediates the asymmetric localization of PAR-3. Development 122: 3133-3140.
2. Qiu, R.G., et al. 2000. A human homolog of the *C. elegans* polarity determinant PAR-6 links Rac and Cdc42 to PKCζ signaling and cell transformation. Curr. Biol. 10: 697-707.
3. Kim, S.K. 2000. Cell polarity: new PARtners for Cdc42 and Rac. Nat. Cell Biol. 2: E143-E145.
4. Joberty, G., et al. 2000. The cell-polarity protein PAR6 links PAR3 and atypical protein kinase C to Cdc42. Nat. Cell Biol. 2: 531-539.
5. Lin, D., et al. 2000. A mammalian PAR-3-PAR-6 complex implicated in Cdc42/Rac1 and aPKC signaling and cell polarity. Nat. Cell Biol. 2: 540-547.
6. Brazil, D.P., et al. 2000 Cell polarity: scaffold proteins par excellence. Curr. Biol. 10: R592-R594.

CHROMOSOMAL LOCATION

Genetic locus: Pard6b (mouse) mapping to 2 H3.

PRODUCT

PARD6B 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PARD6B shRNA Plasmid (m): sc-62752-SH and PARD6B shRNA (m) Lentiviral Particles: sc-62752-V as alternate gene silencing products.

For independent verification of PARD6B (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-62752A, sc-62752B and sc-62752C.

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

PARD6B siRNA (m) is recommended for the inhibition of PARD6B 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 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

PARD6B (B-10): sc-166405 is recommended as a control antibody for monitoring of PARD6B 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 PARD6B gene expression knockdown using RT-PCR Primer: PARD6B (m)-PR: sc-62752-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.