SANTA CRUZ BIOTECHNOLOGY, INC.

DOCK 180 siRNA (m): sc-35208

BACKGROUND

The v-Crk oncogene product shares homologous amino acid sequences, designated Src homology region 2 (SH2) and SH3, with many molecules involved in signal transduction. The v-Crk cellular homolog, c-Crk, is a member of a newly emerging class of genes including Nck and GRB2/ASH which encode proteins that consist primarily of SH2 and SH3 domains. Two distinct human c-Crk cDNAs, designated Crk I and Crk II, have been identified and shown to represent alternative splice products of c-Crk. The major translational product of c-Crk I has been identified as a variably expressed protein, while c-Crk II encodes a widely expressed protein and a more variably expressed protein. The major c-Crk transforming activity appears associated with c-Crk I p28 expression. DOCK 180, a protein downstream of Crk, has been identified as a major Crk-associated protein. When DOCK 180 is recruited to the plasma membrane from a cytoplasmic reservoir, presumably by Crk, changes in cellular morphology and spindle formation occur, suggesting DOCK 180 to be a Crk effector molecule.

REFERENCES

1. Mayer, B.J. and Hanafusa, H. 1990. Association of the v-Crk oncogene product with phosphotyrosine-containing proteins and protein kinase activity. Proc. Natl. Acad. Sci. USA 87: 2638-2642.
2. Matsuda, M., et al. 1990. Binding of transforming protein, P47GAG-Crk, to a broad range of phosphotyrosine-containing proteins. Science 248: 1537-1539.

CHROMOSOMAL LOCATION

Genetic locus: Dock1 (mouse) mapping to 7 F3.

PRODUCT

DOCK 180 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 DOCK 180 shRNA Plasmid (m): sc-35208-SH and DOCK 180 shRNA (m) Lentiviral Particles: sc-35208-V as alternate gene silencing products.

For independent verification of DOCK 180 (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-35208A, sc-35208B and sc-35208C.

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.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

APPLICATIONS

DOCK 180 siRNA (m) is recommended for the inhibition of DOCK 180 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

DOCK 180 (E-2): sc-514080 is recommended as a control antibody for monitoring of DOCK 180 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 DOCK 180 gene expression knockdown using RT-PCR Primer: DOCK 180 (m)-PR: sc-35208-PR (20 µl, 539 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Cote, J.F., et al. 2005. A novel and evolutionarily conserved PtdIns (3,4,5) P_3-binding domain is necessary for DOCK 180 signalling. Nat. Cell Biol. 7: 797-807.
2. Lee, W.L., et al. 2007. Role of Crk II in Fcγ receptor-mediated phagocytosis. J. Biol. Chem. 282: 11135-11143.
3. Mohan, J.J., et al. 2018. Silencing of dedicator of cytokinesis (DOCK 180) obliterates pregnancy by interfering with decidualization due to blockage of nuclear entry of autoimmune regulator (AIRE). Am. J. Reprod. Immunol. 80: e12844.

PROTOCOLS

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