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SANTA CRUZ BIOTECHNOLOGY, INC.

Crk I/II siRNA (h): sc-43704

BACKGROUND

The Crk family of adapter proteins including Crk I, Crk II and Crk-L consist mostly of SH2 and SH3 domains. Through the interactions between SH2 domain and phosphotyrosine residues and/or between SH3 domain and proline-rich motifs, they are involved in a variety of signaling cascades. Crk I and Crk II are encoded by the same gene, which undergoes alternative splicing to yield these two proteins, but differ in their biological activities. Crk II has less transforming activity than Crk I, although both Crk I and Crk II bind to many tyrosine-phosphorylated proteins that bind to GRB2. In addition, Crk II becomes rapidly tyrosine-phosphorylated in response to stimulation with Insulin-like growth factor-I (IGF-I) and might be involved in the IGF-I receptor signaling pathway. The gene encoding Crk I and II maps to human chromosome 17p13.3, a region which demonstrates frequent deletion or loss of heterozygosity in a wide range of human cancers.

REFERENCES

1. Matsuda, M., Tanaka, S., Nagata, S., Kojima, A., Kurata, T. and Shibuya, M. 1992. Two species of human CRK cDNA encode proteins with distinct biological activities. Mol. Cell. Biol. 12: 3482-3489.
2. Fioretos, T., Heisterkamp, N., Groffen, J., Benjes, S. and Morris, C. 1993. CRK proto-oncogene maps to human chromosome band 17p13. Oncogene 10: 2853-2855.
3. Koval, A.P., Blakesley, V.A., Roberts, C.T., Jr., Zick, Y. and Leroith, D. 1998. Interaction *in vitro* of the product of the c-Crk II proto-oncogene with the Insulin-like growth factor-I receptor. Biochem. J. 330: 923-932
4. Imaizumi, T., Araki, K., Miura, K., Araki, M., Suzuki, M., Terasaki, H. and Yamamura, K. 1999. Mutant mice lacking Crk II caused by the gene trap insertional mutagenesis: Crk II is not essential for embryonic development. Biochem. Biophys. Res. Commun. 266: 569-574.
5. SWISS-PROT/TrEMBL (1169096). World Wide Web URL: http://www.expasy.ch/sprot/sprot-top.html

CHROMOSOMAL LOCATION

Genetic locus: CRK (human) mapping to 17p13.3.

PRODUCT

Crk I/II siRNA (h) is a pool of 4 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 Crk I/II shRNA Plasmid (h): sc-43704-SH and Crk I/II shRNA (h) Lentiviral Particles: sc-43704-V as alternate gene silencing products.

For independent verification of Crk I/II (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-43704A, sc-43704B, sc-43704C and sc-43704D.

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 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

Crk I/II siRNA (h) is recommended for the inhibition of Crk I/II 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

Crk II (B-4): sc-390132 is recommended as a control antibody for monitoring of Crk I/II 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 Crk I/II gene expression knockdown using RT-PCR Primer: Crk I/II (h)-PR: sc-43704-PR (20 µl, 425 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.