



Cortactin siRNA (m): sc-35094

BACKGROUND

Cortactin (also designated Ems-1) is a filamentous Actin (F-Actin) binding protein that has been shown to be a substrate for Src p60. Cortactin contains tandem 37 amino acid repeats at the amino-terminus and an SH3 domain at the carboxy-terminus. The tandem repeats appear to be necessary for F-Actin binding. Tyrosine phosphorylation of Cortactin by Src p60 results in diminished F-Actin binding to Cortactin and reduced F-Actin cross-linking activity. Cortactin has also been shown to be phosphorylated in response to FGF-1. Cortactin exhibits abundant expression in megakaryocytes and platelets, and it may play a role in the maturation of megakaryocytes.

REFERENCES

1. Wu, H., et al. 1993. Cortactin, an 80/85-kilodalton pp60^{src} substrate, is a filamentous Actin-binding protein enriched in the cell cortex. *J. Cell Biol.* 120: 1417-1426.
2. Zhan, X., et al. 1993. Murine cortactin is phosphorylated in response to fibroblast growth factor-1 on tyrosine residues late in the G₁ phase of the BALB/c 3T3 cell cycle. *J. Biol. Chem.* 268: 24427-24431.
3. Zhan, X., et al. 1994. Association of fibroblast growth factor receptor-1 with c-Src correlates with association between c-Src and cortactin. *J. Biol. Chem.* 269: 20221-20224.
4. Okamura, H., et al. 1995. p80/85 Cortactin associates with the Src SH2 domain and colocalizes with v-Src in transformed cells. *J. Biol. Chem.* 270: 26613-26618.
5. Huang, C., et al. 1997. Down-regulation of the filamentous Actin cross-linking activity of cortactin by Src-mediated tyrosine phosphorylation. *J. Biol. Chem.* 272: 13911-13915.
6. Zhan, X., et al. 1997. Upregulation of Cortactin expression during the maturation of megakaryocytes. *Blood* 89: 457-464.

CHROMOSOMAL LOCATION

Genetic locus: Cttb (mouse) mapping to 7 F5.

PRODUCT

Cortactin 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 Cortactin shRNA Plasmid (m): sc-35094-SH and Cortactin shRNA (m) Lentiviral Particles: sc-35094-V as alternate gene silencing products.

For independent verification of Cortactin (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-35094A, sc-35094B and sc-35094C.

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

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

Cortactin (H-5): sc-55579 is recommended as a control antibody for monitoring of Cortactin 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 Cortactin gene expression knockdown using RT-PCR Primer: Cortactin (m)-PR: sc-35094-PR (20 μ l, 568 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.