

CCDC49 siRNA (m): sc-142117

BACKGROUND

Spliceosomes are large ribonucleoproteins that remove introns from nuclear pre-mRNA in a two-step reaction. CCDC49 (coiled-coil domain-containing protein 49), also known as Pre-mRNA-splicing factor CWC25 homolog, is a 425 amino acid protein that is involved in the catalytic steps of splicing in *Saccharomyces cerevisiae*. CCDC49 associates with a component of the Prp19-associated complex, CEF1, which is involved in spliceosome activation. It is likely that CCDC49 facilitates juxtaposition of the 5' splice site and branch point during the final step in the first catalytic reaction. There are two isoforms of CCDC49 that are produced as a result of alternative splicing events.

REFERENCES

1. Lamond, A.I. 1993. The spliceosome. *Bioessays* 15: 595-603.
2. Umen, J.G. and Guthrie, C. 1995. The second catalytic step of pre-mRNA splicing. *RNA* 1: 869-885.
3. Tarn, W.Y. and Steitz, J.A. 1997. Pre-mRNA splicing: the discovery of a new spliceosome doubles the challenge. *Trends Biochem. Sci.* 22: 132-137.
4. Nagai, K., Muto, Y., Pomeranz Krummel, D.A., Kambach, C., Ignjatovic, T., Walke, S. and Kuglstatter, A. 2001. Structure and assembly of the spliceosomal snRNPs. *Novartis Medal Lecture. Biochem. Soc. Trans.* 29: 15-26.
5. Nilsen, T.W. 2003. The spliceosome: the most complex macromolecular machine in the cell? *Bioessays* 25: 1147-1149.
6. Chiu, Y.F., Liu, Y.C., Chiang, T.W., Yeh, T.C., Tseng, C.K., Wu, N.Y. and Cheng, S.C. 2009. Cwc25 is a novel splicing factor required after Prp2 and Yju2 to facilitate the first catalytic reaction. *Mol. Cell. Biol.* 29: 5671-5678.
7. Warkocki, Z., Odenwälder, P., Schmitzová, J., Platzmann, F., Stark, H., Urlaub, H., Ficner, R., Fabrizio, P. and Lührmann, R. 2009. Reconstitution of both steps of *Saccharomyces cerevisiae* splicing with purified spliceosomal components. *Nat. Struct. Mol. Biol.* 16: 1237-1243.

CHROMOSOMAL LOCATION

Genetic locus: Cwc25 (mouse) mapping to 11 D.

PRODUCT

CCDC49 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 CCDC49 shRNA Plasmid (m): sc-142117-SH and CCDC49 shRNA (m) Lentiviral Particles: sc-142117-V as alternate gene silencing products.

For independent verification of CCDC49 (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-142117A, sc-142117B and sc-142117C.

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

CCDC49 siRNA (m) is recommended for the inhibition of CCDC49 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CCDC49 gene expression knockdown using RT-PCR Primer: CCDC49 (m)-PR: sc-142117-PR (20 μ l). 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.