

# CNOT6 siRNA (h): sc-72944

## BACKGROUND

CNOT6 is a widely expressed subunit of the CCR4-NOT transcription complex. The CCR4-NOT complex is an evolutionarily conserved, multi-component complex known to be involved in transcription as well as mRNA degradation. Various subunits (e.g. CNOT1, CNOT3) are involved in influencing nuclear hormone receptor activities. The CCR4-NOT complex is also involved in the regulation of Histone H3 lysine 4 methylation through a ubiquitin-dependent pathway that likely involves the proteasome. CNOT6 belongs to the CCR4/nocturin family and contains three LRR (leucine-rich) repeats. In the cytoplasm, CNOT6 acts as a poly(A) nuclease involved in mRNA decay mediated by the major-protein-coding determinant of instability (mCRD) of the Fos gene.

## REFERENCES

1. Albert, T.K., et al. 2000. Isolation and characterization of human orthologs of yeast CCR4-NOT complex subunits. *Nucleic Acids Res.* 28: 809-817.
2. Chen, J., et al. 2002. CCR4, a 3'-5' poly(A) RNA and ssDNA exonuclease, is the catalytic component of the cytoplasmic deadenylase. *EMBO J.* 21: 1414-1426.
3. Dupressoir, A., et al. 2003. Identification of four families of  $\gamma$ CCR4- and  $Mg^{2+}$ -dependent endonuclease-related proteins in higher eukaryotes, and characterization of orthologs of  $\gamma$ CCR4 with a conserved leucine-rich repeat essential for hCAF1/hPOP2 binding. *BMC Genomics* 2: 9.
4. Semotok, J.L., et al. 2005. Smaug recruits the CCR4/POP2/NOT deadenylase complex to trigger maternal transcript localization in the early *Drosophila* embryo. *Curr. Biol.* 15: 284-294.
5. Oh, J.H., et al. 2005. Transcriptome analysis of human gastric cancer. *Mamm. Genome* 16: 942-954.
6. Behm-Ansmant, I., et al. 2006. mRNA degradation by miRNAs and GW182 requires both CCR4-NOT deadenylase and DCP1:DCP2 decapping complexes. *Genes Dev.* 20: 1885-1898.

## CHROMOSOMAL LOCATION

Genetic locus: CNOT6 (human) mapping to 5q35.3.

## PRODUCT

CNOT6 siRNA (h) 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CNOT6 shRNA Plasmid (h): sc-72944-SH and CNOT6 shRNA (h) Lentiviral Particles: sc-72944-V as alternate gene silencing products.

For independent verification of CNOT6 (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-72944A, sc-72944B and sc-72944C.

## RESEARCH USE

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

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

CNOT6 siRNA (h) is recommended for the inhibition of CNOT6 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  $\mu$ M in 66  $\mu$ 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

CNOT6 (2193C2a): sc-81231 is recommended as a control antibody for monitoring of CNOT6 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).

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CNOT6 gene expression knockdown using RT-PCR Primer: CNOT6 (h)-PR: sc-72944-PR (20  $\mu$ l, 533 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Sanchez-Perez, I., et al. 2009. hCCR4/cNOT6 targets DNA-damage response proteins. *Cancer Lett.* 273: 281-291.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.