

# CGNL1 siRNA (h): sc-90135

## BACKGROUND

Diffusion of solutes is prevented across certain barriers by the formation of tight junction seals. Occludin and Cingulin interact with other proteins to direct the formation and regulation of tight junctions. Cingulin, a protein component of the submembrane plaque of tight junctions (TJ), contains globular and coiled-coil domains and interacts *in vitro* with several TJ and cytoskeletal proteins, including the PDZ protein ZO-1. CGNL1 (cingulin-like 1), also known as JACOP (junction-associated coiled-coil protein), is a 1,302 amino acid tight junction protein belonging to the Cingulin family. Expressed in smooth muscle, spleen, testis, fetal brain, amygdala, corpus callosum, cerebellum, thalamus and subthalamic nucleus of adult brain. CGNL1 exists as two alternatively spliced isoforms and is encoded by a gene located on human chromosome 15q21.3. CGNL1 may participate in anchoring the apical junctional complex, primarily tight junctions, to actin-based cytoskeletons. Mutations in the gene encoding CGNL1 is the cause of aromatase excess syndrome, which is characterized by an estrogen excess due to an increased aromatase activity.

## REFERENCES

1. D'Atri, F. et al. 2001. Cingulin interacts with F-Actin *in vitro*. FEBS Lett. 507: 21-24.
2. D'Atri, F. et al. 2002. Evidence for a functional interaction between cingulin and ZO-1 in cultured cells. J. Biol. Chem. 277: 27757-27764.
3. Shozu, M., et al. 2003. Estrogen excess associated with novel gain-of-function mutations affecting the aromatase gene. N. Engl. J. Med. 348: 1855-1865.
4. Bordin, M., et al. 2004. Histone deacetylase inhibitors up-regulate the expression of tight junction proteins. Mol. Cancer Res. 2: 692-701.
5. Guillemot, L., et al. 2004. Disruption of the cingulin gene does not prevent tight junction formation but alters gene expression. J. Cell Sci. 117: 5245-5256.

## CHROMOSOMAL LOCATION

Genetic locus: CGNL1 (human) mapping to 15q21.3.

## PRODUCT

CGNL1 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 CGNL1 shRNA Plasmid (h): sc-90135-SH and CGNL1 shRNA (h) Lentiviral Particles: sc-90135-V as alternate gene silencing products.

For independent verification of CGNL1 (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-90135A, sc-90135B and sc-90135C.

## PROTOCOLS

See our web site at [www.scbt.com](http://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  $\mu$ 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

CGNL1 siRNA (h) is recommended for the inhibition of CGNL1 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

CGNL1 (E-6): sc-377525 is recommended as a control antibody for monitoring of CGNL1 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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 $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 CGNL1 gene expression knockdown using RT-PCR Primer: CGNL1 (h)-PR: sc-90135-PR (20  $\mu$ 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.