## SANTA CRUZ BIOTECHNOLOGY, INC.

# KLHL25 siRNA (h): sc-89948



## BACKGROUND

KLHL25 (ectoderm-neural cortex protein 2, ENC2) is a cytoplasmic protein that contains six kelch regions and a single BTB (POZ) domain. KLHL25 is highly homologus to another Kelch-like protein, ENC1, and it is believed to operate in a manner similar to other Kelch-domain containing proteins. Kelchdomain repeat containing proteins often act as modifiers of Actin fibers. Expressed early in embryogenesis, ENC1 helps to mediate neuronal process formation. It also appears to have a role in neural crest cell differentiation. KLHL25 likely functions as a substrate specific adapter for protein ubiquitinating complexes. KLHL25 is expressed in most tissues with highest expression in brain and liver.

## REFERENCES

- Hernandez, M.C., et al. 1997. ENC-1: a novel mammalian kelch-related gene specifically expressed in the nervous system encodes an Actin-binding protein. J. Neurosci. 17: 3038-3051.
- Seng, S., et al. 2006. KLHL1/MRP2 mediates neurite outgrowth in a glycogen synthase kinase 3β-dependent manner. Mol. Cell. Biol. 26: 8371-8384.
- 3. Peeters, T., et al. 2006. Kelch-repeat proteins interacting with the  $G_{\alpha}$  protein Gpa2 bypass adenylate cyclase for direct regulation of protein kinase A in yeast. Proc. Natl. Acad. Sci. USA 103: 13034-13039.
- Angers, S., et al. 2006. The KLHL12-Cullin-3 ubiquitin ligase negatively regulates the Wnt-β-catenin pathway by targeting dishevelled for degradation. Nat. Cell Biol. 8: 348-357.
- 5. Peeters, T., et al. 2007. Directly from  $G_\alpha$  to protein kinase A: the kelch repeat protein bypass of adenylate cyclase. Trends Biochem. Sci. 32: 547-554.
- 6. Niranjan, T., et al. 2007. Kelch repeat protein interacts with the yeast  $G_{\alpha}$  subunit Gpa2p at a site that couples receptor binding to guanine nucleotide exchange. J. Biol. Chem. 282: 24231-24238.
- Nacak, T.G., et al. 2007. The BTB-Kelch protein KLEIP controls endothelial migration and sprouting angiogenesis. Circ. Res. 100: 1155-1163.
- Aromolaran, K.A., et al. 2007. The Kelch-like protein 1 modulates P/Q-type calcium current density. Neuroscience 145: 841-850.

#### CHROMOSOMAL LOCATION

Genetic locus: KLHL25 (human) mapping to 15q25.3.

#### PRODUCT

KLHL25 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 KLHL25 shRNA Plasmid (h): sc-89948-SH and KLHL25 shRNA (h) Lentiviral Particles: sc-89948-V as alternate gene silencing products.

For independent verification of KLHL25 (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-89948A, sc-89948B and sc-89948C.

#### 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**

KLHL25 siRNA (h) is recommended for the inhibition of KLHL25 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

KLHL25 (K-20): sc-100774 is recommended as a control antibody for monitoring of KLHL25 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<sup>®</sup> 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<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

#### **RT-PCR REAGENTS**

Semi-quantitative RT-PCR may be performed to monitor KLHL25 gene expression knockdown using RT-PCR Primer: KLHL25 (h)-PR: sc-89948-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.

## PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.