

## KLHL25 siRNA (m): sc-146525

### 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 homologous to another Kelch-like protein, ENC1, and it is believed to operate in a manner similar to other Kelch-domain containing proteins. Kelch-domain 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 3 $\beta$ -dependent manner. *Mol. Cell. Biol.* 26: 8371-8384.
- 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- $\beta$ -catenin pathway by targeting dishevelled for degradation. *Nat. Cell Biol.* 8: 348-357.
- 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.
- 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.
- Nacac, 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.
- Rondou, P., et al. 2008. BTB Protein KLHL12 targets the dopamine D4 receptor for ubiquitination by a Cul3-based E3 ligase. *J. Biol. Chem.* 283: 11083-11096.

### CHROMOSOMAL LOCATION

Genetic locus: Klhl25 (mouse) mapping to 7 D2.

### PRODUCT

KLHL25 siRNA (m) is a target-specific 19-25 nt siRNA 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 (m): sc-146525-SH and KLHL25 shRNA (m) Lentiviral Particles: sc-146525-V as alternate gene silencing 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

KLHL25 siRNA (m) is recommended for the inhibition of KLHL25 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  $\mu$ 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-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 KLHL25 gene expression knockdown using RT-PCR Primer: KLHL25 (m)-PR: sc-146525-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](http://www.scbt.com) for detailed protocols and support products.