

# KLHL13 siRNA (h): sc-91014

## BACKGROUND

KLHL13 (kelch-like 13), also known as BKLHD2 (BTB and kelch domain-containing protein 2), is a 604 amino acid protein that contains six Kelch repeats and one BTB/POZ domain. Expressed predominantly in brain, KLHL13 is believed to play a role in protein ubiquitination and may function as a substrate-specific adapter of an E3 ubiquitin-protein ligase complex. E3 ligases accept a ubiquitin residue from an E2 ubiquitin-conjugating enzyme and immediately transfer that residue to a protein that is targeted for degradation. Specifically, KLHL13 interacts with KLHL9 and CUL-3, a member of the cullin family of mediators that participate in the selective targeting of proteins for ubiquitin-mediated proteolysis. Via its BTB and C-terminal Kelch (BACK) motif, KLHL13 is thought to play a role in spatially orientating substrates in the CUL-3 ligase.

## REFERENCES

1. Singer, J.D., et al. 1999. Cullin-3 targets cyclin E for ubiquitination and controls S phase in mammalian cells. *Genes Dev.* 13: 2375-2387.
2. Tyers, M. and Willems, A.R. 1999. One ring to rule a superfamily of E3 ubiquitin ligases. *Science* 284: 601, 603-604.
3. Nagase, T., et al. 2000. Prediction of the coding sequences of unidentified human genes. XVI. The complete sequences of 150 new cDNA clones from brain which code for large proteins *in vitro*. *DNA Res.* 7: 65-73.
4. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 300655. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Morrow, C.J., et al. 2005. Bub1 and aurora B cooperate to maintain BubR1-mediated inhibition of APC/CCdc20. *J. Cell Sci.* 118: 3639-3652.
6. Sumara, I. and Peter, M. 2007. A Cul3-based E3 ligase regulates mitosis and is required to maintain the spindle assembly checkpoint in human cells. *Cell Cycle* 6: 3004-3010.
7. Sumara, I., et al. 2007. A Cul3-based E3 ligase removes aurora B from mitotic chromosomes, regulating mitotic progression and completion of cytokinesis in human cells. *Dev. Cell* 12: 887-900.

## CHROMOSOMAL LOCATION

Genetic locus: KLHL13 (human) mapping to Xq24.

## PRODUCT

KLHL13 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 KLHL13 shRNA Plasmid (h): sc-91014-SH and KLHL13 shRNA (h) Lentiviral Particles: sc-91014-V as alternate gene silencing products.

For independent verification of KLHL13 (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-91014A, sc-91014B and sc-91014C.

## 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

KLHL13 siRNA (h) is recommended for the inhibition of KLHL13 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

KLHL9/13 (D-4): sc-166486 is recommended as a control antibody for monitoring of KLHL13 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 KLHL13 gene expression knockdown using RT-PCR Primer: KLHL13 (h)-PR: sc-91014-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## PROTOCOLS

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