

## KLHL5 siRNA (h): sc-89298

### BACKGROUND

KLHL5, also known as kelch-like protein 5, is a 755 amino acid protein similar to the *Drosophila* kelch protein. Localized to the cytoskeleton, KLHL5 is expressed at higher levels in thyroid gland, adrenal gland and ovary and has lower expression in spinal cord, prostate, lymph node, trachea and testis. KLHL5 contains six kelch repeats and one BTB (POZ) domain. The BTB (broad complex, tramtrack and bric-a-brac) domain, also known as the POZ (poxvirus and zinc finger) domain, is an N-terminal homodimerization domain that contains multiple copies of kelch repeats and/or C<sub>2</sub>H<sub>2</sub>-type zinc fingers. Proteins that contain BTB domains are thought to be involved in transcriptional regulation via control of chromatin structure and function. KLHL5 is expressed as three isoforms produced by alternative splicing.

### REFERENCES

1. Albagli, O., et al. 1995. The BTB/POZ domain: a new protein-protein interaction motif common to DNA- and actin-binding proteins. *Cell Growth Differ.* 6: 1193-1198.
2. Robinson, D.N. and Cooley, L. 1997. *Drosophila* kelch is an oligomeric ring canal actin organizer. *J. Cell Biol.* 138: 799-810.
3. Melnick, A., et al. 2000. In-depth mutational analysis of the promyelocytic leukemia zinc finger BTB/POZ domain reveals motifs and residues required for biological and transcriptional functions. *Mol. Cell. Biol.* 20: 6550-6567.
4. Adams, J., et al. 2000. The kelch repeat superfamily of proteins: propellers of cell function. *Trends Cell Biol.* 10: 17-24.
5. Wang, S., et al. 2001. Cloning and characterization of KLHL5, a novel human gene encoding a kelch-related protein with a BTB domain. *Biochem. Genet.* 39: 227-238.
6. Online Mendelian Inheritance in Man, OMIM<sup>™</sup>. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 608064. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
7. Prag, S. and Adams, J.C. 2003. Molecular phylogeny of the kelch-repeat superfamily reveals an expansion of BTB/kelch proteins in animals. *BMC Bioinformatics* 4: 42.
8. Geyer, R., et al. 2003. BTB/POZ domain proteins are putative substrate adaptors for cullin 3 ubiquitin ligases. *Mol. Cell* 12: 783-790.

### CHROMOSOMAL LOCATION

Genetic locus: KLHL5 (human) mapping to 4p14.

### PRODUCT

KLHL5 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 KLHL5 shRNA Plasmid (h): sc-89298-SH and KLHL5 shRNA (h) Lentiviral Particles: sc-89298-V as alternate gene silencing products.

For independent verification of KLHL5 (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-89298A, sc-89298B and sc-89298C.

### 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

KLHL5 siRNA (h) is recommended for the inhibition of KLHL5 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

KLHL5 (A-2): sc-515745 is recommended as a control antibody for monitoring of KLHL5 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<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> 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<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 KLHL5 gene expression knockdown using RT-PCR Primer: KLHL5 (h)-PR: sc-89298-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

### SELECT PRODUCT CITATIONS

1. Schleifer, R.J., et al. 2018. KLHL5 knockdown increases cellular sensitivity to anticancer drugs. *Oncotarget* 9: 37429-37438.

### RESEARCH USE

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