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# SIAH-1/2 siRNA (h): sc-44102

## BACKGROUND

SIAH, the human homolog of the *Drosophila* seven in absentia (*sina*) gene, is a tumor suppressor protein that is expressed in intestinal epithelium and activated during apoptosis. Human SIAH proteins are produced as two distinct gene products, SIAH-1, and the slightly larger protein SIAH-2, which share a highly conserved C-terminal sequence and differ in their N-terminal regions. SIAH-1 contains an N-terminal RING-finger domain, which is required for proteolysis, and a cystein-rich C-terminal domain, which regulates oligomerization and SIAH binding to target proteins. As a tumor suppressor, SIAH-1 binds DCC (deleted in colorectal cancer) and regulates DCC degradation via the ubiquitin-proteasome pathway. SIAH-1 also binds a Bcl-2 related protein, Bag-1, thereby inhibiting cell growth. The majority of SIAH-1 is localized to the nucleus, however a small percentage is detected in the cytoplasm. This nuclear localization suggests that SIAH proteins may interact with other nuclear matrix proteins and DNA.

## REFERENCES

1. Nemani, M., et al. 1996. Activation of the human homologue of the *Drosophila* *sina* gene in apoptosis and tumor suppression. *Proc. Natl. Acad. Sci. USA* 93: 9039-9042.
2. Hu, G., et al. 1997. Mammalian homologs of seven in absentia regulate DCC via the ubiquitin-proteasome pathway. *Genes Dev.* 11: 2701-2714.
3. Hu, G., et al. 1997. Characterization of human homologs fo the *Drosophila* seven in absentia (*sina*) gene. *Genomics* 46: 103-111.
4. Matsuzawas, S., et al. 1998. P53-inducible human homologue of *Drosophila* seven in absentia (*Siah*) inhibits cell growth: suppression by BAG-1. *EMBO J.* 17: 2736-2747.
5. Hu, G., et al. 1999. Siah-1 N-terminal RING domain is required for proteolysis function, and C-terminal sequence regulate oligomerization and binding to target proteins. *Mol. Cell. Biol.* 19: 724-732.
6. Roperch, J., et al. 1999. SIAH-1 promotes apoptosis and tumor suppression through a network involving the regulation of protein folding, unfolding, and trafficking: identification of common effectors with p53 and p21Waf1. *Proc. Natl. Acad. Sci. USA* 96: 8070-8073.

## CHROMOSOMAL LOCATION

Genetic locus: SIAH1 (human) mapping to 16q12.1, SIAH2 (human) mapping to 3q25.1.

## PRODUCT

SIAH-1/2 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 SIAH-1/2 shRNA Plasmid (h): sc-44102-SH and SIAH-1/2 shRNA (h) Lentiviral Particles: sc-44102-V as alternate gene silencing products.

For independent verification of SIAH-1/2 (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-44102A, sc-44102B and sc-44102C.

## 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

SIAH-1/2 siRNA (h) is recommended for the inhibition of SIAH-1/2 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

SIAH-1/2 (8G7H12): sc-81785 is recommended as a control antibody for monitoring of SIAH-1/2 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).

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor SIAH-1 gene expression knockdown using RT-PCR Primer: SIAH-1/2 (h)-PR: sc-44102-PR (20  $\mu$ l, 584 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Krämer, O.H., et al. 2008. Mechanism for ubiquitylation of the leukemia fusion proteins AML1-ETO and PML-RAR $\alpha$ . *FASEB J.* 22: 1369-1379.
2. Buchwald, M., et al. 2012. SIAH ubiquitin ligases target the nonreceptor tyrosine kinase ACK1 for ubiquitinylation and proteasomal degradation. *Oncogene* 32: 4913-4920.

## 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.