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# HAH1 siRNA (h): sc-45252

## BACKGROUND

Delivery of copper to a specific P-type ATPase in the Golgi apparatus (Ccc2 in yeast, ATP7A and ATP7B in humans) is an important step in copper homeostasis that is accomplished by a small copper chaperone protein. HAH1 (also designated ATOX1), a metal transport protein that belongs to the ATX1 family, is involved in cellular antioxidant defense and can bind and deliver cytosolic copper to the copper ATPase proteins. Both HAH1 (the human homolog of Atx1) and Atx1 preferentially interact with domains 2 and 4 of ATP7B. Atx1 also interacts with both Ccc2 domains.

## REFERENCES

1. Klomp, L.W., et al. 1997. Identification and functional expression of HAH1, a novel human gene involved in copper homeostasis. *J. Biol. Chem.* 272: 9221-9226.
2. Harrison, M.D., et al. 2000. Intracellular copper routing: the role of copper chaperones. *Trends Biochem. Sci.* 25: 29-32.
3. Boultonwood, J., et al. 2000. Physical mapping of the human ATX1 homologue (HAH1) to the critical region of the 5q-syndrome within 5q32, and immediately adjacent to the SPARC gene. *Hum. Genet.* 106: 127-129.
4. van Dongen, E.M., et al. 2004. Copper-dependent protein-protein interactions studied by yeast two-hybrid analysis. *Biochem. Biophys. Res. Commun.* 323: 789-795.
5. Anastassopoulou, I., et al. 2004. Solution structure of the apo and copper(I)-loaded human metallochaperone HAH1. *Biochemistry* 43: 13046-13053.

## CHROMOSOMAL LOCATION

Genetic locus: ATOX1 (human) mapping to 5q33.1.

## PRODUCT

HAH1 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 HAH1 shRNA Plasmid (h): sc-45252-SH and HAH1 shRNA (h) Lentiviral Particles: sc-45252-V as alternate gene silencing products.

For independent verification of HAH1 (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-45252A, sc-45252B and sc-45252C.

## STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at  $-20^{\circ}$  C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at  $-20^{\circ}$  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

HAH1 siRNA (h) is recommended for the inhibition of HAH1 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

HAH1 (D-9): sc-398742 is recommended as a control antibody for monitoring of HAH1 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 HAH1 gene expression knockdown using RT-PCR Primer: HAH1 (h)-PR: sc-45252-PR (20  $\mu$ l). Annealing temperature for the primers should be  $55-60^{\circ}$  C and the extension temperature should be  $68-72^{\circ}$  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.