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# HAX-1 shRNA (m) Lentiviral Particles: sc-43366-V

## BACKGROUND

HAX-1 (HS1-associated protein X-1 or HS1-binding protein protein X-1), encodes a novel protein. HAX-1 has previously been shown to associate with HS1, a protein specifically expressed in cells of the hematopoietic lineage, and is thought to be involved in signal transduction in B cells and apoptosis. Though first identified as a protein that associates with HS1, recent data has also revealed interactions between HAX-1 and three disparate proteins: Polycystin-2 (derived from the PKD2 gene), a protein linked to polycystic kidney disease; Cortactin; and EBNA-LP (Epstein-Barr virus nuclear antigen leader protein). Additionally, HAX-1 has been identified as a binding partner to the carboxy-terminus of the K15 protein of Kaposi's sarcoma-associated herpesvirus. K15 interacts with cellular HAX-1 *in vitro* and *in vivo*. Furthermore, HAX-1 co-localizes with K15 in the endoplasmic reticulum and mitochondria. Immunofluorescence experiments show that in most cells PKD2 and HAX-1 co-localize in the cell body, but in some cells PKD2 and HAX-1 also are sorted into cellular processes and lamellipodia. The HAX-1 gene is expressed ubiquitously among tissues. Its protein is localized mainly in mitochondria, but also in endoplasmic reticulum and the nuclear envelope of the cell.

## REFERENCES

1. Suzuki, Y., et al. 1997. HAX-1, a novel intracellular protein, localized on mitochondria, directly associates with HS1, a substrate of Src family tyrosine kinases. *J. Immunol.* 158: 2736-2744.
2. Gallagher, A.R., et al. 2000. The polycystic kidney disease protein PKD2 interacts with HAX-1, a protein associated with the Actin cytoskeleton. *Proc. Natl. Acad. Sci. USA* 97: 4017-4022.
3. Kawaguchi, Y., et al. 2000. Interaction of Epstein-Barr virus nuclear antigen leader protein (EBNA-LP) with HS1-associated protein X-1: implication of cytoplasmic function of EBNA-LP. *J. Virol.* 74: 10104-10111.

## CHROMOSOMAL LOCATION

Genetic locus: Hax1 (mouse) mapping to 3 F1.

## PRODUCT

HAX-1 shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 2 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 µl frozen stock containing  $1.0 \times 10^6$  infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see HAX-1 siRNA (m): sc-43366 and HAX-1 shRNA Plasmid (m): sc-43366-SH as alternate gene silencing products.

## STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

## PROTOCOLS

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

## APPLICATIONS

HAX-1 shRNA (m) Lentiviral Particles is recommended for the inhibition of HAX-1 expression in mouse cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing  $1.0 \times 10^6$  infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

## GENE EXPRESSION MONITORING

HAX-1 (FL-279): sc-28268 is recommended as a control antibody for monitoring of HAX-1 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor HAX-1 gene expression knockdown using RT-PCR Primer: HAX-1 (m)-PR: sc-43366-PR (20 µl, 538 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

## RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.