

# RHBDL1 siRNA (h): sc-93492

## BACKGROUND

Members of the rhomboid family of integral membrane proteins are related to *Drosophila* Rhomboid-1, a serine protease that cleaves the membrane domain of the *Drosophila* EGF-family protein, Spitz, to release a soluble growth factor. Rhomboid-related protein 1 (RHBDL1) is a 438 amino acid multi-pass membrane protein belonging to the rhomboid family. As an intramembrane serine protease, RHBDL1 cleaves type-1 transmembrane domains using a catalytic dyad composed of serine and histidine that are contributed by different transmembrane domains. Believed to be expressed in heart, brain, skeletal muscle and kidney, RHBDL1 can exist as two isoforms due to alternative splicing events. RHBDL1 is encoded by a gene mapping to human chromosome 16p13.3.

## REFERENCES

- Urban, S., Lee, J.R. and Freeman, M. 2001. *Drosophila* rhomboid-1 defines a family of putative intramembrane serine proteases. *Cell* 107: 173-182.
- Urban, S., Lee, J.R. and Freeman, M. 2002. A family of rhomboid intramembrane proteases activates all *Drosophila* membrane-tethered EGF ligands. *EMBO J.* 21: 4277-4286.
- Urban, S. and Freeman, M. 2003. Substrate specificity of rhomboid intramembrane proteases is governed by helix-breaking residues in the substrate transmembrane domain. *Mol. Cell* 11: 1425-1434.
- Pascall, J.C. and Brown, K.D. 2004. Intramembrane cleavage of ephrinB3 by the human rhomboid family protease, RHBDL2. *Biochem. Biophys. Res. Commun.* 317: 244-252.
- Online Mendelian Inheritance in Man, OMIM™. 2004. Johns Hopkins University, Baltimore, MD. MIM Number: 608962. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
- Urban, S. 2006. Rhomboid proteins: conserved membrane proteases with divergent biological functions. *Genes Dev.* 20: 3054-3068.

## CHROMOSOMAL LOCATION

Genetic locus: RHBDL1 (human) mapping to 16p13.3.

## PRODUCT

RHBDL1 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 RHBDL1 shRNA Plasmid (h): sc-93492-SH and RHBDL1 shRNA (h) Lentiviral Particles: sc-93492-V as alternate gene silencing products.

For independent verification of RHBDL1 (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-93492A, sc-93492B and sc-93492C.

## PROTOCOLS

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

## 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

RHBDL1 siRNA (h) is recommended for the inhibition of RHBDL1 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

RHBDL1 (5B11): sc-517160 is recommended as a control antibody for monitoring of RHBDL1 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 RHBDL1 gene expression knockdown using RT-PCR Primer: RHBDL1 (h)-PR: sc-93492-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## RESEARCH USE

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