

# UNC5H4 siRNA (h): sc-63187

## BACKGROUND

The UNC5H family of proteins act as transmembrane receptors for netrin-1 and play a crucial role in axon guidance and migration of neural cells. Additionally, when cleaved by a caspase to produce an intracellular fragment containing a death domain, UNC5H receptors induce apoptosis. This activity is blocked by the binding of netrin-1. In the absence of netrin-1, UNC5H receptors act as tumor suppressors by inhibiting anchorage-independent growth and invasion, but mutation of these receptors provides a potential mechanism for tumorigenicity. The expression of UNC5H receptors is down-regulated in multiple carcinomas, including colorectal, breast, ovary, uterus, stomach, lung, and kidney cancers. UNC5H4, also known as UNC5D (unc-5 homolog D), is single-pass type I membrane protein that is a member of the UNC5H netrin receptor family. Two isoforms of UNC5H4 exist due to alternative splicing events.

## REFERENCES

1. Llambi, F., et al. 2001. Netrin-1 acts as a survival factor via its receptors Unc5H and DCC. *EMBO J.* 20: 2715-2722.
2. Komatsuzaki, K., et al. 2002. Modulation of G<sub>i</sub>  $\alpha$ 2 signaling by the axonal guidance molecule Unc5H2. *Biochem. Biophys. Res. Commun.* 297: 898-905.
3. Thiebault, K., et al. 2003. The netrin-1 receptors Unc5H are putative tumor suppressors controlling cell death commitment. *Proc. Natl. Acad. Sci. USA* 100: 4173-4178.
4. Kruger, R.P., et al. 2004. Mapping netrin receptor binding reveals domains of Unc5 regulating its tyrosine phosphorylation. *J. Neurosci.* 24: 10826-10834.
5. Kuramoto, T., et al. 2004. Rat neurological mutations cerebellar vermis defect and hobble are caused by mutations in the netrin-1 receptor gene Unc5h3. *Brain Res. Mol. Brain Res.* 122: 103-108.
6. Klar, J., et al. 2005. RAR-related orphan receptor A isoform 1 (RORa1) is disrupted by a balanced translocation t(4;15)(q22.3;q21.3) associated with severe obesity. *Eur. J. Hum. Genet.* 13: 928-934.
7. Dillon, A.K., et al. 2007. Unc5C is required for spinal accessory motor neuron development. *Mol. Cell. Neurosci.* 35: 482-489.

## CHROMOSOMAL LOCATION

Genetic locus: UNC5D (human) mapping to 8p12.

## PRODUCT

UNC5H4 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 UNC5H4 shRNA Plasmid (h): sc-63187-SH and UNC5H4 shRNA (h) Lentiviral Particles: sc-63187-V as alternate gene silencing products.

For independent verification of UNC5H4 (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-63187A, sc-63187B and sc-63187C.

## 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

UNC5H4 siRNA (h) is recommended for the inhibition of UNC5H4 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

UNC5H4 (4G3): sc-517085 is recommended as a control antibody for monitoring of UNC5H4 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 UNC5H4 gene expression knockdown using RT-PCR Primer: UNC5H4 (h)-PR: sc-63187-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.

## PROTOCOLS

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