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# Notch 1 shRNA (h2) Lentiviral Particles: sc-44226-V

## BACKGROUND

The LIN-12/Notch family of transmembrane receptors is believed to play a central role in development by regulating cell fate decisions. To date, four Notch homologs have been identified in mammals and have been designated Notch 1, Notch 2, Notch 3 and Notch 4. The Notch genes are expressed in a variety of tissues in both the embryonic and adult organism, suggesting that the genes are involved in multiple signaling pathways. The Notch proteins have been found to be overexpressed or rearranged in human tumors. Ligands for Notch include Jagged1, Jagged2 and Delta. Jagged can activate Notch and prevent myoblast differentiation by inhibiting the expression of muscle regulatory and structural genes. Jagged2 is thought to be involved in the development of various tissues whose development is dependent upon epithelial-mesenchymal interactions. Normal Delta expression is restricted to the adrenal gland and placenta. Delta expression has also been found in neuroendocrine tumors such as neuroblastomas and pheochromocytomas.

## REFERENCES

- Weinmaster, G., et al. 1992. Notch 2: a second mammalian Notch gene. *Development* 116: 931-941.
- Kopan, R., et al. 1993. Mouse Notch: expression in hair follicles correlates with cell fate determination. *J. Cell Biol.* 121: 631-641.
- Laborda, J., et al. 1993. dIk, a putative mammalian homeotic gene differentially expressed in small cell lung carcinomas and neuroendocrine tumor cell line. *J. Biol. Chem.* 268: 3817-3820.
- Swiatek, P.J., et al. 1994. Notch 1 is essential for postimplantation development in mice. *Genes Dev.* 8: 707-719.
- Lindsell, C.E., et al. 1995. Jagged: a mammalian ligand that activates Notch 1. *Cell* 80: 909-917.
- Uyttendaele, H., et al. 1996. Notch 4/int-3, a mammary proto-oncogene, is an endothelial cell-specific mammalian Notch gene. *Development* 122: 2251-2259.

## CHROMOSOMAL LOCATION

Genetic locus: NOTCH1 (human) mapping to 9q34.3.

## PRODUCT

Notch 1 shRNA (h2) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200  $\mu$ l 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 Notch 1 siRNA (h2): sc-44226 and Notch 1 shRNA Plasmid (h2): sc-44226-SH as alternate gene silencing products.

## STORAGE

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

## APPLICATIONS

Notch 1 shRNA (h2) Lentiviral Particles is recommended for the inhibition of Notch 1 expression in human cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200  $\mu$ l 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

Notch 1 (A-8): sc-376403 is recommended as a control antibody for monitoring of Notch 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-mouse IgM-HRP: sc-2064 (dilution range: 1:500-1:5,000), TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgM-FITC: sc-2082 (dilution range: 1:100-1:400) or goat anti-mouse IgM-TR: sc-2983 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Notch 1 gene expression knockdown using RT-PCR Primer: Notch 1 (h2)-PR: sc-44226-PR (20  $\mu$ l, 467 bp). Annealing temperature for the primers should be  $55-60^\circ$  C and the extension temperature should be  $68-72^\circ$  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.

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