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# Gemin3 shRNA (h) Lentiviral Particles: sc-43798-V

## BACKGROUND

Spinal muscular atrophy (SMA) is an autosomal recessive neurodegenerative disease characterized by loss of motor neurons in the spinal cord. SMA is caused by deletion or loss-of-function mutations in the SMN (survival of motor neuron) gene. Gemin3, also known as DP103, DDX20, DEAD-box protein DP130 and DEAD/H box 20, is a protein product of human chromosome 1p13.2. It associates directly with SMN and is a part of the SMN complex containing Gemin2, Gemin4, Gemin5 and Gemin6, as well as several spliceosomal snRNP proteins. The SMN complex plays an essential role in spliceosomal snRNP assembly in the cytoplasm and is required for pre-mRNA splicing of the nucleus. It is found in both the cytoplasm and the nucleus. The nuclear form is concentrated in subnuclear bodies called gems (for Gemini of the coiled bodies). Gemin3 also interacts with SmB, Smd2 and Smd3. It contains the conserved motif Asp-Glu-Ala-Asp (DEAD) characteristic of DEAD-box proteins. Gemin3 is a putative RNA helicase and shows ATPase activity. It is expressed in B and T cell neuroblastoma-derived cell lines, malignant melanoma tumor, normal testis and is expressed in low levels in colon, skeletal muscle, liver, kidney and lung.

## REFERENCES

1. Fischer, U., et al. 1997. The SMN-SIP1 complex has an essential role in spliceosomal snRNP biogenesis. *Cell* 90: 1023-1029.
2. Coover, D., et al. 1997. The survival motor neuron protein in spinal muscular atrophy. *Hum. Mol. Genet.* 6: 1205-1214.
3. Monani, U., et al. 1999. A single nucleotide difference that alters splicing patterns distinguishes the SMA gene SMN1 from the copy gene SMN2. *Hum. Mol. Genet.* 8: 1177-1183.

## CHROMOSOMAL LOCATION

Genetic locus: DDX20 (human) mapping to 1p13.2.

## PRODUCT

Gemin3 shRNA (h) Lentiviral Particles are concentrated, transduction-ready viral particles containing a target-specific construct that encodes a 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 Gemin3 siRNA (h): sc-43798 and Gemin3 shRNA Plasmid (h): sc-43798-SH as alternate gene silencing products.

## STORAGE

Store lentiviral particles at  $-80^\circ\text{C}$ . Stable for at least one year from the date of shipment. Once thawed, particles can be stored at  $4^\circ\text{C}$  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

Gemin3 shRNA (h) Lentiviral Particles is recommended for the inhibition of Gemin3 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

Gemin3 (12H12): sc-57007 is recommended as a control antibody for monitoring of Gemin3 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 IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Gemin3 gene expression knockdown using RT-PCR Primer: Gemin3 (h)-PR: sc-43798-PR (20  $\mu$ l, 492 bp). Annealing temperature for the primers should be  $55-60^\circ\text{C}$  and the extension temperature should be  $68-72^\circ\text{C}$ .

## 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.