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Nucleostemin shRNA (m) Lentiviral Particles: sc-45831-V

BACKGROUND

Nucleostemin, also designated nucleolar GTP-binding protein 3, is a member of the MMR1/HSR1 GTP-binding protein family. It is expressed in the nucleoli of adult CNS stem cells, primitive bone marrow cells, embryonic stem cells and in several cancer cell lines. It is often considered a stem cell marker. Over-expression or depletion of the protein can reduce cell proliferation in CNS stem cells. Nucleostemin shuttles between the nucleus and the nucleolus and may be important in maintaining the proliferative capacity of stem cells. It is important in the growth regulation of liver cancer, gastric cancer and several other cancer types. The gene encoding for Nucleostemin is localized to chromosome 3p21.1.

REFERENCES

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2. Normile, D., et al. 2002. Cell proliferation. Common control for cancer, stem cells. *Science* 298: 1869.
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4. Schwartz, P.H., et al. 2003. Isolation and characterization of neural progenitor cells from post-mortem human cortex. *J. Neurosci. Res.* 74: 838-851.
5. Baddoo, M., et al. 2003. Characterization of mesenchymal stem cells isolated from murine bone marrow by negative selection. *J. Cell Biochem.* 89: 1235-1249.
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CHROMOSOMAL LOCATION

Genetic locus: Gnl3 (mouse) mapping to 14 B.

PRODUCT

Nucleostemin shRNA (m) 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 μ l frozen stock containing 1.0×10^6 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 Nucleostemin siRNA (m): sc-45831 and Nucleostemin shRNA Plasmid (m): sc-45831-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.

APPLICATIONS

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

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 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

Nucleostemin (F-5): sc-398978 is recommended as a control antibody for monitoring of Nucleostemin 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 Nucleostemin gene expression knockdown using RT-PCR Primer: Nucleostemin (m)-PR: sc-45831-PR (20 μ l, 581 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.

PROTOCOLS

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