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Reptin 52 shRNA (h) Lentiviral Particles: sc-43544-V

BACKGROUND

Pontin 52 is a nuclear matrix protein that is primarily expressed in the nucleus and is also present in the cytoplasm. Pontin 52 is expressed in the nucleoplasm of whole cells, but is not present in the nucleoli. Pontin 52 is also designated RUVBL1, for *E. coli* RuvB-like 1 protein, or NMP 238, and is the human homolog of rat TIP49. Pontin 52 contains an ATPase/helicase motif and may represent a class of cofactors recruited by transcriptional activation domains that function in diverse pathways. For instance, *in vivo*, Pontin 52 is complexed with Myc and Reptin 52, which is a Pontin 52 related protein also designated RUVBL2. The interaction of Pontin 52 with Myc is dependent upon a Myc domain essential for oncogenic activity, suggesting that functional Pontin 52 is an essential mediator of Myc oncogenic transformation.

REFERENCES

- Holzmann, K., et al. 1998. Identification and characterization of the ubiquitously occurring nuclear matrix protein NMP 238. *Biochem. Biophys. Res. Commun.* 252: 39-45.
- Bauer, A., et al. 1998. Pontin 52, an interaction partner of β -catenin, binds to the TATA box-binding protein. *Proc. Natl. Acad. Sci. USA* 95: 14787-14792.
- Qiu, X.B., et al. 1998. An eukaryotic RuvB-like protein (RUVBL1) essential for growth. *J. Biol. Chem.* 273: 27786-27793.
- Makino, Y., et al. 1998. TIP49, homologous to the bacterial DNA helicase RuvB, acts as an autoantigen in human. *Biochem. Biophys. Res. Commun.* 245: 819-823.
- Wood, M.A., et al. 2000. An ATPase/helicase complex is an essential cofactor for oncogenic transformation by c-Myc. *Mol. Cell* 5: 321-330.

CHROMOSOMAL LOCATION

Genetic locus: RUVBL2 (human) mapping to 19q13.33.

PRODUCT

Reptin 52 shRNA (h) 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 Reptin 52 siRNA (h): sc-43544 and Reptin 52 shRNA Plasmid (h): sc-43544-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.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

APPLICATIONS

Reptin 52 shRNA (h) Lentiviral Particles is recommended for the inhibition of Reptin 52 expression in human 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

Reptin 52 (B-5): sc-374135 is recommended as a control antibody for monitoring of Reptin 52 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 Reptin 52 gene expression knockdown using RT-PCR Primer: Reptin 52 (h)-PR: sc-43544-PR (20 μ l). 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.