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Parafibromin shRNA (m) Lentiviral Particles: sc-45529-V

BACKGROUND

Parathyroid tumors are heterogeneous and diagnosis of the disease is often difficult. The Parafibromin protein may be important as a marker for diagnosing parathyroid carcinoma. Parafibromin is encoded by the endocrine tumor suppressor gene CDC73 (cell division cycle 73, Paf1/RNA polymerase II complex component), alternatively known as the HRPT2 (hyperparathyroidism-jaw tumor syndrome 2) gene. The human CDC73 gene, which maps to chromosome 1q25, is the human homolog of *Saccharomyces cerevisiae* Cdc73 and is re-sponsible for the hyperparathyroidism with jaw tumor syndrome (HPT-JT). Parafibromin is part of the RNA polymerase II/Paf1 complex, which is crucial for histone modification. This Parafibromin complex binds to both the nonphosphorylated forms and the Ser 2- and Ser 5-phosphorylated forms of the RNA polymerase II large subunit.

REFERENCES

1. Simonds, W.F., et al. 2004. Familial isolated hyperparathyroidism is rarely caused by germline mutation in HRPT2, the gene for the hyperparathyroidism-jaw tumor syndrome. *J. Clin. Endocrinol. Metab.* 89: 96-102.
2. Cavaco, B.M., et al. 2004. Hyperparathyroidism-jaw tumor syndrome in Roma families from Portugal is due to a founder mutation of the HRPT2 gene. *J. Clin. Endocrinol. Metab.* 89: 1747-1752.
3. Haven, C.J., et al. 2004. Gene expression of parathyroid tumors: molecular subclassification and identification of the potential malignant phenotype. *Cancer Res.* 64: 7405-7411.
4. Tan, M.H., et al. 2004. Loss of parafibromin immunoreactivity is a distinguishing feature of parathyroid carcinoma. *Clin. Cancer Res.* 10: 6629-6637.
5. Cetani, F., et al. 2004. Genetic analyses of the HRPT2 gene in primary hyperparathyroidism: germline and somatic mutations in familial and sporadic parathyroid tumors. *J. Clin. Endocrinol. Metab.* 89: 5583-5591.
6. Rozenblatt-Rosen, O., et al. 2005. The parafibromin tumor suppressor protein is part of a human Paf1 complex. *Mol. Cell. Biol.* 25: 612-620.

CHROMOSOMAL LOCATION

Genetic locus: Cdc73 (mouse) mapping to 1 F.

PRODUCT

Parafibromin 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 Parafibromin siRNA (m): sc-45529 and Parafibromin shRNA Plasmid (m): sc-45529-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

Parafibromin shRNA (m) Lentiviral Particles is recommended for the inhibition of Parafibromin 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

Parafibromin (G-11): sc-365053 is recommended as a control antibody for monitoring of Parafibromin 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 Parafibromin gene expression knockdown using RT-PCR Primer: Parafibromin (m)-PR: sc-45529-PR (20 μ l, 532 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.

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