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Pumilio 2 shRNA (h) Lentiviral Particles: sc-44773-V

BACKGROUND

Pumilio 2 is a sequence-specific RNA-binding protein that regulates translation and mRNA stability by binding mRNA targets. It supports proliferation and self-renewal of stem cells by regulating the translation of key transcripts. The Pumilio gene encodes proteins that are required for development of germ stem cells in one or both sexes. The Pumilio protein interacts with the human Nanos1 protein, and this interaction may play a conserved role in germ cell development. Pumilio 2 is highly expressed in testis and ovary and at lower levels in brain, heart, kidney, liver, muscle, placenta, intestine and stomach. It is also expressed in stem cells, germ cells and in most fetal tissues.

REFERENCES

1. Spassov, D.S. and Jurecic, R. 2002. Cloning and comparative sequence analysis of PUM1 and PUM2 genes, human members of the Pumilio family of RNA-binding proteins. *Gene* 299: 195-204
2. Jaruzelska, J., et al. 2003. Conservation of a Pumilio-Nanos complex from *Drosophila* germ plasm to human germ cells. *Dev. Genes Evol.* 213: 120-126
3. Moore, F.L., et al. 2003. Human Pumilio 2 is expressed in embryonic stem cells and germ cells and interacts with DAZ (deleted in azoospermia) and DAZ-like proteins. *Proc. Natl. Acad. Sci. USA* 100: 538-543
4. Vessey, J.P., et al. 2006. Dendritic localization of the translational repressor Pumilio 2 and its contribution to dendritic stress granules. *J. Neurosci.* 26: 6496-6508.
5. Spik, A., et al. 2006. Candidate mRNAs interacting with fertility protein PUMILIO2 in the human germ line. *Reprod. Biol.* 1:37-42.
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CHROMOSOMAL LOCATION

Genetic locus: PUM2 (human) mapping to 2p24.1.

PRODUCT

Pumilio 2 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 Pumilio 2 siRNA (h): sc-44773 and Pumilio 2 shRNA Plasmid (h): sc-44773-SH as alternate gene silencing products.

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.

APPLICATIONS

Pumilio 2 shRNA (h) Lentiviral Particles is recommended for the inhibition of Pumilio 2 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

Pumilio 2 (C-8): sc-514108 is recommended as a control antibody for monitoring of Pumilio 2 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 Pumilio 2 gene expression knockdown using RT-PCR Primer: Pumilio 2 (h)-PR: sc-44773-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° 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.

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.