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Exo1 shRNA (h) Lentiviral Particles: sc-44880-V

BACKGROUND

Comparative evaluation of the expression patterns of the human and mouse genes, combined with previous biochemical and yeast genetic studies, indicate that the Exo1 (Exonuclease I) proteins are important contributors to chromosome processing during mammalian DNA repair and recombination. In mice, the Exo1 gene maps to distal chromosome 1, consistent with the recent mapping of the orthologous human HEX1/EXO1 gene to chromosome 1q43. Exo1 is expressed prominently in testis, an area of active homologous recombination, and spleen, a prominent lymphoid tissue. In both mammalian and yeast systems, Exo1 is a 5'-3' double stranded DNA exonuclease that has previously been implicated in DNA mismatch repair (MMR). The MMR system ensures genome integrity by removing mispaired and unpaired bases that originate during replication. In humans, Exo1 interacts with MSH2 and MLH1 and has been proposed to be a redundant exonuclease in MMR. In both mammalian and yeast systems, Exo1 plays a structural role in MMR and stabilizes multiprotein complexes containing a number of MMR proteins.

REFERENCES

1. Lee, B.I., et al. 1999. Expression specificity of the mouse exonuclease 1 (mExo1) gene. *Nucleic Acids Res.* 27: 4114-4120.
2. Kirkpatrick, D.T., et al. 2000. Decreased meiotic intergenic recombination and increased meiosis I nondisjunction in exo1 mutants of *Saccharomyces cerevisiae*. *Genetics* 156: 1549-1557.
3. Tran, P.T., Simon, J.A. and Liskay, R.M. 2001. Interactions of Exo1p with components of MutL α in *Saccharomyces cerevisiae*. *Proc. Natl. Acad. Sci. USA* 98: 9760-9765.
4. Mansour, A.A., et al. 2001. Control of GT repeat stability in *Schizosaccharomyces pombe* by mismatch repair factors. *Genetics* 158: 77-85.
5. Amin, N.S., et al. 2001. exo1-Dependent mutator mutations: model system for studying functional interactions in mismatch repair. *Mol. Cell. Biol.* 21: 5142-5155.

CHROMOSOMAL LOCATION

Genetic locus: EXO1 (human) mapping to 1q43.

PRODUCT

Exo1 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 Exo1 siRNA (h): sc-44880 and Exo1 shRNA Plasmid (h): sc-44880-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

Exo1 shRNA (h) Lentiviral Particles is recommended for the inhibition of Exo1 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

Exo1 (H-300): sc-33194 is recommended as a control antibody for monitoring of Exo1 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-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (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-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Exo1 gene expression knockdown using RT-PCR Primer: Exo1 (h)-PR: sc-44880-PR (20 μ l, 565 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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