

OSTM1 siRNA (m): sc-151337

BACKGROUND

OSTM1 (osteopetrosis associated transmembrane protein 1), also known as gl (gray-lethal) or HSPC019, is a 338 amino acid single-pass type I membrane protein that is expressed primarily in osteoclasts and melanocytes as well as brain, kidney and spleen. Bone autosomal recessive osteopetrosis (ARO) is the most severe form of hereditary bone disease whose cellular basis is in the osteoclast and is characterized by abnormally dense bone, due to defective resorption of immature bone. ARO is suggested to be caused by mutations in the OSTM1 gene. The disorder occurs in two forms: a severe autosomal recessive form occurring in utero, infancy, or childhood, and a benign autosomal dominant form occurring in adolescence or adulthood. Defects in the OSTM1 gene are also the cause of the spontaneous gl mutant, which is responsible for a coat color defect in mice.

REFERENCES

1. Rajapurohitam, V., et al. 2001. The mouse osteopetrotic grey-lethal mutation induces a defect in osteoclast maturation/function. *Bone* 28: 513-523.
2. Chalhoub, N., et al. 2003. Grey-lethal mutation induces severe malignant autosomal recessive osteopetrosis in mouse and human. *Nat. Med.* 9: 399-406.
3. Ramírez, A., et al. 2004. Identification of a novel mutation in the coding region of the grey-lethal gene OSTM1 in human malignant infantile osteopetrosis. *Hum. Mutat.* 23: 471-476.
4. Blin-Wakkach, C., et al. 2004. Osteopetrosis, from mouse to man. *Med. Sci.* 20: 61-67.
5. Pangrazio, A., et al. 2006. Mutations in OSTM1 (grey lethal) define a particularly severe form of autosomal recessive osteopetrosis with neural involvement. *J. Bone Miner. Res.* 21: 1098-1105.
6. Lange, P.F., et al. 2006. CLC-7 requires OSTM1 as a β -subunit to support bone resorption and lysosomal function. *Nature* 440: 220-223.

CHROMOSOMAL LOCATION

Genetic locus: *Ostm1* (mouse) mapping to 10 B2.

PRODUCT

OSTM1 siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see OSTM1 shRNA Plasmid (m): sc-151337-SH and OSTM1 shRNA (m) Lentiviral Particles: sc-151337-V as alternate gene silencing products.

For independent verification of OSTM1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-151337A, sc-151337B and sc-151337C.

PROTOCOLS

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

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

OSTM1 siRNA (m) is recommended for the inhibition of OSTM1 expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

OSTM1 (4H1): sc-293366 is recommended as a control antibody for monitoring of OSTM1 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 reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor OSTM1 gene expression knockdown using RT-PCR Primer: OSTM1 (m)-PR: sc-151337-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.