

OSX siRNA (m): sc-45909

BACKGROUND

Osterix (OSX) is a zinc finger-containing transcriptional activator that is distinctly expressed in all developing bones and is important for osteoblast differentiation. In particular, OSX is implicated in the differentiation of osteoblasts, which are the specialized cells of bone formation. OSX is a nuclear protein that binds to GC box promoters elements and activates mRNA synthesis from genes containing functional recognition sites. The periosteal and mesenchymal cells of the membranous skeletal elements of OSX⁻ mice fail to differentiate into osteoblasts. Subsequently, the mesenchymal cells of OSX⁻ mice fail to deposit bone matrix and do not form bone. Cox-2 deficiency correlates with a decrease in OSX expression, suggesting that Cox-2 may induce OSX to mediate skeletal repair.

REFERENCES

1. Nakashima, K., et al. 2002. The novel zinc finger-containing transcription factor osterix is required for osteoblast differentiation and bone formation. *Cell* 108: 17-29.
2. Yagi, K., et al. 2003. Bone morphogenetic protein-2 enhances osterix gene expression in chondrocytes. *J. Cell. Biochem.* 88: 1077-1083.
3. Lee, M.H., et al. 2003. BMP-2-induced osterix expression is mediated by Dlx-5 but is independent of RUNX2. *Biochem. Biophys. Res. Commun.* 309: 689-694.
4. Huang, L., et al. 2004. Expression of preosteoblast markers and Cbfa-1 and osterix gene transcripts in stromal tumour cells of giant cell tumour of bone. *Bone* 34: 393-401.

CHROMOSOMAL LOCATION

Genetic locus: Sp7 (mouse) mapping to 15 F3.

PRODUCT

OSX siRNA (m) is a pool of 2 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 OSX shRNA Plasmid (m): sc-45909-SH and OSX shRNA (m) Lentiviral Particles: sc-45909-V as alternate gene silencing products.

For independent verification of OSX (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-45909A and sc-45909B.

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

OSX shRNA Plasmid (m) is recommended for the inhibition of OSX 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

OSX (F-3): sc-393325 is recommended as a control antibody for monitoring of OSX 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 OSX gene expression knockdown using RT-PCR Primer: OSX (m)-PR: sc-45909-PR (20 μ l, 399 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Yano, H., et al. 2014. Sp7/Osterix induces the mouse pro- α 2(I) collagen gene (Col1a2) expression via the proximal promoter in osteoblastic cells. *Biochem. Biophys. Res. Commun.* 452: 531-536.
2. Varanasi, V.G., et al. 2016. Enhanced osteoprogenitor elongated collagen fiber matrix formation by bioactive glass ionic silicon dependent on Sp7 (osterix) transcription. *J. Biomed. Mater. Res. A* 104: 2604-2615.

RESEARCH USE

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

PROTOCOLS

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