

Amelogenin siRNA (m): sc-44846

BACKGROUND

Dental enamel is a highly mineralized tissue with most of its volume occupied by large, highly organized hydroxyapatite crystals. This structure is thought to be controlled through the interaction of many organic matrix molecules including Amelogenin, Ameloblastin, Enamelin, Tuftelin and several other enzymes. All of these secreted proteins are involved in the mineralization and enamel matrix formation in developing tooth enamel. The gene AMELX which encodes for the protein Amelogenin, is encoded on the X-chromosome. Amelogenin, also designated AMG, AMGX or AMEX, is involved in biomineralization and organization of developing enamel. It functions by regulating crystallite formation during the secretory stage of enamel development. Amelogenin, which localizes to the extracellular matrix, is expressed by ameloblasts and is the predominant protein in developing dental enamel. Defects in the AMELX gene can cause amelogenesis imperfecta hypoplastic type 1 (AIH1) which is an X-linked disease that affects the formation of tooth enamel.

REFERENCES

1. Nakahori, Y., et al. 1991. A human X-Y homologous region encodes "Amelogenin". *Genomics* 9: 264-269.
2. Catalano-Sherman, J., et al. 1993. Amino acid sequence of a major human Amelogenin protein employing Edman degradation and cDNA sequencing. *J. Dent. Res.* 72: 1566-1572.
3. Catalano-Sherman, J., et al. 1994. Production of a monoclonal antibody against human amelogenin. *Calcif. Tissue Int.* 54: 76-80.
4. Lagerstrom-Fermer, M., et al. 1995. Amelogenin signal peptide mutation: correlation between mutations in the amelogenin gene (AMGX) and manifestations of X-linked amelogenesis imperfecta. *Genomics* 26: 159-162.
5. Du, C., et al. 2005. Apatite/Amelogenin coating on titanium promotes osteogenic gene expression. *J. Dent. Res.* 84: 1070-1074.
6. Foster, B.L., et al. 2005. Regenerating the periodontium: is there a magic formula? *Orthod. Craniofac. Res.* 8: 285-291.
7. Torres-Quintana, M.A., et al. 2005. Ameloblastin and amelogenin expression in postnatal developing mouse molars. *J. Oral Sci.* 47: 27-34.
8. Wang, H., et al. 2005. Enamel matrix protein interactions. *J. Bone Miner. Res.* 20: 1032-1040.

CHROMOSOMAL LOCATION

Genetic locus: Amelx (mouse) mapping to X F5.

PRODUCT

Amelogenin 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 Amelogenin shRNA Plasmid (m): sc-44846-SH and Amelogenin shRNA (m) Lentiviral Particles: sc-44846-V as alternate gene silencing products.

For independent verification of Amelogenin (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-44846A, sc-44846B and sc-44846C.

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

Amelogenin siRNA (m) is recommended for the inhibition of Amelogenin 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

Amelogenin (F-11): sc-365284 is recommended as a control antibody for monitoring of Amelogenin 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 Amelogenin gene expression knockdown using RT-PCR Primer: Amelogenin (m)-PR: sc-44846-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.

PROTOCOLS

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