SANTA CRUZ BIOTECHNOLOGY, INC.

myogenin siRNA (h): sc-29402

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The Power to Question

BACKGROUND

Differentiation of myogenic cells is regulated by multiple positively and negatively acting factors. One well characterized family of helix-loop-helix (HLH) proteins known to play an important role in the regulation of muscle cell development includes Myo D, myogenin, Myf-5 and Myf-6 (also designated MRF-4 or herculin). Of interest, most muscle cells express either Myo D or Myf-5 in the committed state, but when induced to differentiate, all turn on expression of myogenin. Myo D transcription factors form heterodimers with products of a more widely expressed family of bHLH genes, the E family, which consists of at least three distinct genes: E2A, IF2 and HEB. Myo D-E heterodimers bind avidly to consensus (CANNTG) E box target sites that are functionally important elements in the upstream regulatory sequences of many muscle-specific terminal differentiation genes.

REFERENCES

1. Wright, W.E., et al. 1989. Myogenin, a factor regulating myogenesis, has a domain homologs to Myo D. Cell 56: 607-617.
2. Braun, T., et al. 1989. A novel human muscle factor related to but distinct from Myo D1 induces myogenic conversion in 10T1/2 fibroblasts. EMBO J. 8: 701-709.
3. Rhodes, S.J., et al. 1989. Identification of MRF4: a new member of the muscle regulatory factor gene family. Genes Dev. 3: 2050-2061.
4. Miner, J.H., et al. 1990. Herculin, a fourth member of the Myo D family of myogenic regulatory genes. Proc. Natl. Acad. Sci. USA 87: 1089-1093.
5. Braun, T., et al. 1990. Myf-6, a new member of the human gene family of myogenic determination factors: evidence for a gene cluster on chromosome 12. EMBO J. 9: 821-831.
6. Hollenberg, S.M., et al. 1993. Use of a conditional Myo D transcription factor in studies of Myo D *trans*-activation and muscle determination. Proc. Natl. Acad. Sci. USA 90: 8028-8032.
7. Neuhold, L.A., et al. 1993. HLH forced dimers: tethering Myo D to E47 generates a dominant positive myogenic factor insulated from negative regulation by Id. Cell 74: 1033-1042.

CHROMOSOMAL LOCATION

Genetic locus: MYOG (human) mapping to 1q32.1.

PRODUCT

myogenin siRNA (h) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see myogenin shRNA Plasmid (h): sc-29402-SH and myogenin shRNA (h) Lentiviral Particles: sc-29402-V as alternate gene silencing products.

For independent verification of myogenin (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-29402A, sc-29402B and sc-29402C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

myogenin siRNA (h) is recommended for the inhibition of myogenin expression in human 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 µM in 66 µl. 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

myogenin (5FD): sc-52903 is recommended as a control antibody for monitoring of myogenin 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 myogenin gene expression knockdown using RT-PCR Primer: myogenin (h)-PR: sc-29402-PR (20 µl, 491 bp). 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.