SANTA CRUZ BIOTECHNOLOGY, INC.

Id2 siRNA (m): sc-38001

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The Power to Question

BACKGROUND

Members of the Id family of basic helix-loop-helix (bHLH) proteins include Id1, Id2, Id3 and Id4. They are ubiquitously expressed and dimerize with members of the class A and B HLH proteins. Due to the absence of the basic region, the resulting heterodimers cannot bind DNA. The Id-type proteins thus appear to negatively regulate DNA binding of bHLH proteins. Since Id1 inhibits DNA binding of E12 and MyoD, it apparently functions to inhibit muscle-specific gene expression. Under conditions that facilitate muscle cell differentiation, the Id protein levels fall, allowing E12 and/or E47 to form heterodimers with MyoD and myogenin, which in turn activate myogenic differentiation. It has been shown that expression of each of the Id proteins is strongly dependent on growth factor activation and that reduction of Id mRNA levels by antisense oligonucleotides leads to a delayed reentry of arrested cells into the cell cycle following growth factor stimulation.

REFERENCES

1. Benezra, R., et al. 1990. The protein Id: a negative regulator of helix-loop-helix DNA binding proteins. Cell 61: 49-59.
2. Christy, B.A., et al. 1991. An Id-related helix-loop-helix protein encoded by a growth factor-inducible gene. Proc. Natl. Acad. Sci. USA 88: 1815-1819.
3. Sun, X., et al. 1991. Id proteins Id1 and Id2 selectively inhibit DNA binding by one class of helix-loop-helix proteins. Mol. Cell. Biol. 11: 5603-5611.
4. Neuhold, L.A. and Wold, B. 1993. HLH forced dimers: tethering MyoD to E47 generates a dominant positive myogenic factor insulated from negative regulation by Id. Cell 74: 1033-1042.
5. Riechmann, V., et al. 1994. The expression pattern of Id4, a novel dominant negative helix-loop-helix protein, is distinct from Id1, Id2 and Id3. Nucleic Acids Res. 22: 749-755.
6. Barone, M.V., et al. 1994. Id proteins control growth induction in mammalian cells. Proc. Natl. Acad. Sci. USA 91: 4985-4988.
7. Hara, E., et al. 1994. Id-related genes encoding helix-loop-helix proteins are required for G_1 progression and are repressed in senescent human fibroblasts. J. Biol. Chem. 269: 2139-2145.
8. LocusLink Report (LocusID: 3398). http://www.ncbi.nlm.nih.gov/LocusLink/

CHROMOSOMAL LOCATION

Genetic locus: Id2 (mouse) mapping to 12 A1.3.

PRODUCT

Id2 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Id2 shRNA Plasmid (m): sc-38001-SH and Id2 shRNA (m) Lentiviral Particles: sc-38001-V as alternate gene silencing products.

For independent verification of Id2 (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-38001A, sc-38001B and sc-38001C.

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

Id2 siRNA (m) is recommended for the inhibition of Id2 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 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

Id2 (E-7): sc-398104 is recommended as a control antibody for monitoring of Id2 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Id2 gene expression knockdown using RT-PCR Primer: Id2 (m)-PR: sc-38001-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Chakrabarti, L., et al. 2013. A mechanism linking Id2-TGFβ crosstalk to reversible adaptive plasticity in neuroblastoma. PLoS ONE 8: e83521.
2. DiVito, K.A., et al. 2014. Id2, Id3 and Id4 overcome a Smad7-mediated block in tumorigenesis, generating TGFβ-independent melanoma. Carcinogenesis 35: 951-958.

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.