SANTA CRUZ BIOTECHNOLOGY, INC.

ASH2L siRNA (m): sc-77338

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The Power to Question

BACKGROUND

The human ASH2L gene encodes a 628 amino acid protein known as ASH2L1, or isoform 1, which contains a nuclear localization signal and PHD finger motif, suggesting that the gene product functions as a transcription regulator. Alternative splicing results in a shorter isoform 2, designated ASH2L2, which is missing the first 94 amino acid residues found in ASH2L1. Human ASH2L proteins are 60% homologous to *Drosophila* ash2, which positively regulates expression of certain genes in early development and contain similar, but not identical, domains, including a zinc finger motif. ASH2L is highly expressed in fetal liver, testis and leukemia cell lines with erythroid and megakaryocytic potential, such as K-562, Hel and Dami. Differentiation inducers (e.g. phorbol ester and hemin) cause different expression patterns in these cells lines, suggesting that ASH2L plays a role in hematopoiesis and is associated with particular types of leukemia.

REFERENCES

1. Ikegawa, S., et al. 1999. Cloning and characterization of ASH2L and Ash2l, human and mouse homologs of the *Drosophila* ash2 gene. Cytogenet. Cell Genet. 84: 167-172.
2. Wang, J., et al. 2001. ASH2L: alternative splicing and downregulation during induced megakaryocytic differentiation of multipotential leukemia cell lines. J. Mol. Med. 79: 399-405.
3. Amoros, M., et al. 2002. The ash2 gene is involved in *Drosophila* wing development. Int. J. Dev. Biol. 46: 321-324.
4. Wysocka, J., et al. 2003. Human Sin3 deacetylase and trithorax-related Set1/ash2 histone H3-K4 methyltransferase are tethered together selectively by the cell-proliferation factor HCF-1. Genes Dev. 17: 896-911.
5. Beltran, S., et al. 2003. Transcriptional network controlled by the trithorax-group gene ash2 in *Drosophila melanogaster*. Proc. Natl. Acad. Sci. USA 100: 3293-3298.
6. Angulo, M., et al. 2004. Activation and repression activities of ash2 in *Drosophila* wing imaginal discs. Development 131: 4943-4953.

CHROMOSOMAL LOCATION

Genetic locus: Ash2l (mouse) mapping to 8 A2.

PRODUCT

ASH2L 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 ASH2L shRNA Plasmid (m): sc-77338-SH and ASH2L shRNA (m) Lentiviral Particles: sc-77338-V as alternate gene silencing products.

For independent verification of ASH2L (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-77338A, sc-77338B and sc-77338C.

RESEARCH USE

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

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

ASH2L siRNA (m) is recommended for the inhibition of ASH2L 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

ASH2L (2046D2a): sc-81184 is recommended as a control antibody for monitoring of ASH2L 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 ASH2L gene expression knockdown using RT-PCR Primer: ASH2L (m)-PR: sc-77338-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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