

ASH1L siRNA (h): sc-88574

BACKGROUND

ASH1L (absent small and homeotic disks protein 1 homolog) is a 2,969 amino acid protein encoded by the human gene ASH1L. ASH1L belongs to the histone-lysine methyltransferase family (SET2 subfamily) and contains three AT hook DNA-binding domains, one AWS domain, one BAH domain, one bromo domain, one PHD-type zinc finger, one post-SET domain and one SET domain. It is a widely expressed nuclear protein with highest expression found in brain, heart and kidney. ASH1L is a histone methyltransferase and is believed to methylate Lys 4 of Histone H3, which is a specific tag for epigenetic transcriptional activation.

REFERENCES

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6. Gregory, G.D., Vakoc, C.R., Rozovskaia, T., Zheng, X., Patel, S., Nakamura, T., Canaani, E. and Blobel, G.A. 2007. Mammalian ASH1L is a histone methyltransferase that occupies the transcribed region of active genes. *Mol. Cell. Biol.* 27: 8466-8479.

CHROMOSOMAL LOCATION

Genetic locus: ASH1L (human) mapping to 1q22.

PRODUCT

ASH1L 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ASH1L shRNA Plasmid (h): sc-88574-SH and ASH1L shRNA (h) Lentiviral Particles: sc-88574-V as alternate gene silencing products.

For independent verification of ASH1L (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-88574A, sc-88574B and sc-88574C.

PROTOCOLS

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

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

ASH1L siRNA (h) is recommended for the inhibition of ASH1L 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 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

ASH1L (ASH5H03): sc-81052 is recommended as a control antibody for monitoring of ASH1L 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 ASH1L gene expression knockdown using RT-PCR Primer: ASH1L (h)-PR: sc-88574-PR (20 μ l, 503 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.