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HLTF siRNA (h): sc-45943

BACKGROUND

The protein encoded by the HLTF gene is a member of the SWI/SNF family of proteins. Members of this family have helicase and ATPase activities. They are thought to regulate transcription of certain genes by altering the chromatin structure around those genes. The HLTF encoded protein contains a RING finger DNA-binding motif. Two transcript variants encoding the same protein have been found for this gene. However, use of an alternative translation start site produces an isoform which is truncated at the N-terminus as compared to the full-length protein. Transcriptional inactivation of HLTF by aberrant DNA methylation and histone deacetylation may be involved in stomach carcinogenesis through down-regulation of HLTF expression.

REFERENCES

1. Mansharamani, M., et al. 2001. Cloning and characterization of an atypical type IV P-type ATPase that binds to the RING motif of RUSH transcription factors. *J. Biol. Chem.* 276: 3641-3649.
2. Moinova, H.R., et al. 2002. HLTF gene silencing in human colon cancer. *Proc. Natl. Acad. Sci. USA* 99: 4562-4567.
3. Hamai, Y., et al. 2003. DNA hypermethylation and histone hypoacetylation of the HLTF gene are associated with reduced expression in gastric carcinoma. *Cancer Sci.* 94: 692-698.
4. Leung, W.K., et al. 2003. Inactivation of helicase-like transcription factor by promoter hypermethylation in human gastric cancer. *Mol. Carcinog.* 37: 91-97.
5. SWISS-PROT/TrEMBL (NP_003062). World Wide Web URL: <http://www.expasy.ch/sprot/sprot-top.html>

CHROMOSOMAL LOCATION

Genetic locus: HRLF (human) mapping to 3q24.

PRODUCT

HLTF 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 HLTF shRNA Plasmid (h): sc-45943-SH and HLTF shRNA (h) Lentiviral Particles: sc-45943-V as alternate gene silencing products.

For independent verification of HLTF (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-45943A, sc-45943B and sc-45943C.

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

HLTF shRNA (h) Lentiviral Particles is recommended for the inhibition of HLTF 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

HLTF (G-6): sc-398357 is recommended as a control antibody for monitoring of HLTF 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 HLTF gene expression knockdown using RT-PCR Primer: HLTF (h)-PR: sc-45943-PR (20 μ l). 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.