SANTA CRUZ BIOTECHNOLOGY, INC.

HHAT siRNA (m): sc-145953

BACKGROUND

HHAT (Hedgehog acyltransferase), also known as MART2, SKI1, SIT or SKN, is a 493 amino acid multi-pass membrane protein that localizes to the endoplasmic reticulum and belongs to the membrane-bound acyltransferase family. Expressed ubiquitously, HHAT functions to catalyze the N-terminal palmitoylation of SSH (slingshot homolog), an event that is required for SHH signaling pathways. HHAT is expressed in cancer cell lines, suggesting a role for HHAT in tumorigenesis. The gene encoding HHAT maps to human chromosome 1q32.2 and is expressed as four alternatively spliced isoforms. Chromosome 1 is the largest human chromosome, spanning about 260 million base pairs and making up 8% of the human genome. Several disorders, including Stickler syndrome, Parkinsons disease, Gaucher disease, malignant melanoma and Usher syndrome, are caused by defects in genes that localize to chromosome 1.

REFERENCES

1. Kawakami, Y., Wang, X., Shofuda, T., Sumimoto, H., Tupesis, J., Fitzgerald, E. and Rosenberg, S. 2001. Isolation of a new melanoma antigen, MART-2, containing a mutated epitope recognized by autologous tumor-infiltrating T lymphocytes. J. Immunol. 166: 2871-2877.
2. Chamoun, Z., Mann, R.K., Nellen, D., von Kessler, D.P., Bellotto, M., Beachy, P.A. and Basler, K. 2001. Skinny hedgehog, an acyltransferase required for palmitoylation and activity of the hedgehog signal. Science 293: 2080-2084.
3. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 605743. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Katoh, Y. and Katoh, M. 2005. Hedgehog signaling pathway and gastric cancer. Cancer Biol. Ther. 4: 1050-1054.
5. Bergeron, E., Vincent, M.J. and Nichol, S.T. 2007. Crimean-Congo hemorrhagic fever virus glycoprotein processing by the endoprotease SKI-1/S1P is critical for virus infectivity. J. Virol. 81: 13271-13276.
6. Buglino, J.A. and Resh, M.D. 2008. Hhat is a palmitoylacyltransferase with specificity for N-palmitoylation of Sonic Hedgehog. J. Biol. Chem. 283: 22076-22088.

CHROMOSOMAL LOCATION

Genetic locus: Hhat (mouse) mapping to 1 H6.

PRODUCT

HHAT 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 HHAT shRNA Plasmid (m): sc-145953-SH and HHAT shRNA (m) Lentiviral Particles: sc-145953-V as alternate gene silencing products.

For independent verification of HHAT (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-145953A, sc-145953B and sc-145953C.

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

HHAT siRNA (m) is recommended for the inhibition of HHAT 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor HHAT gene expression knockdown using RT-PCR Primer: HHAT (m)-PR: sc-145953-PR (20 µl). 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.