SANTA CRUZ BIOTECHNOLOGY, INC.

hepcidin siRNA (m): sc-145941

BACKGROUND

Hepcidin, also known as HAMP, HEPC, LEAP1 or HFE2B, is an 84 amino acid secreted protein that regulates iron-related signaling events. Highly expressed in liver with lower expression in heart, brain, lung, prostate and thyroid, hepcidin is thought to maintain iron homeostasis and, in conjunction with the HFE protein (a protein that is defective in hereditary hemochromatosis), may mediate both iron storage in macrophages and intestinal iron absorption. Additionally, hepcidin has strong antimicrobial activity against Gram-positive and Gram-negative bacteria, as well as certain yeast strains, suggesting that hepcidin may play a crucial role in staving off bacterial infections. Defects in the gene encoding hepcidin are the cause of hemochromatosis type 2B (also known as juvenile hemochromatosis), an early-onset autosomal recessive disorder that results in severe iron overload and is characterized by hepatic fibrosis, hypogonadotrophic hypogonadism and cardiomyopathy.

REFERENCES

1. Krause, A., et al. 2000. LEAP-1, a novel highly disulfide-bonded human peptide, exhibits antimicrobial activity. FEBS Lett. 480: 147-150.
2. Park, C.H., et al. 2001. Hepcidin, a urinary antimicrobial peptide synthesized in the liver. J. Biol. Chem. 276: 7806-7810.
3. Klüver, E., et al. 2002. Chemical synthesis of β-defensins and LEAP-1/hepcidin. J. Pept. Res. 59: 241-248.
4. Merryweather-Clarke, A.T., et al. 2003. Digenic inheritance of mutations in HAMP and HFE results in different types of haemochromatosis. Hum. Mol. Genet. 12: 2241-2247.
5. Roetto, A., et al. 2003. Mutant antimicrobial peptide hepcidin is associated with severe juvenile hemochromatosis. Nat. Genet. 33: 21-22.

CHROMOSOMAL LOCATION

Genetic locus: Hamp (mouse) mapping to 7 B1.

PRODUCT

hepcidin siRNA (m) is a target-specific 19-25 nt siRNA 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 hepcidin shRNA Plasmid (m): sc-145941-SH and hepcidin shRNA (m) Lentiviral Particles: sc-145941-V as alternate gene silencing 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 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

hepcidin siRNA (m) is recommended for the inhibition of hepcidin 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 60 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

hepcidin/Hamp2 (K-12): sc-240553 is recommended as a control antibody for monitoring of hepcidin 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor hepcidin gene expression knockdown using RT-PCR Primer: hepcidin (m)-PR: sc-145941-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 or our catalog for detailed protocols and support products.