



Herc4 siRNA (h): sc-90436

BACKGROUND

Ubiquitination is an important mechanism through which three classes of enzymes act in concert to target short-lived or abnormal proteins for destruction. The three classes of enzymes involved in ubiquitination are the ubiquitin-activating enzymes (E1s), the ubiquitin-conjugating enzymes (E2s) and the ubiquitin-protein ligases (E3s). Herc4 (HECT domain and RCC1-like domain-containing protein 4) is a 1,057 amino acid cytoplasmic protein that contains one HECT domain and seven RCC1 repeats. Involved in the pathway of protein modification, Herc4 is thought to function as an E3 ubiquitin-protein ligase that accepts ubiquitin (in the form of a thioester) from an E2 ubiquitin-conjugating enzyme and transfers that ubiquitin residue to substrates targeted for degradation. With highest expression in testis during spermatogenesis, Herc4 is required for proper maturation and removal of the cytoplasmic droplet in order for spermatozoan to become fully functional. There are six isoforms of Herc4 that are produced as a result of alternative splicing events.

REFERENCES

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2. Hochrainer, K., et al. 2005. The human HERC family of ubiquitin ligases: novel members, genomic organization, expression profiling, and evolutionary aspects. *Genomics* 85: 153-164.
3. Online Mendelian Inheritance in Man, OMIM™. 2005. Johns Hopkins University, Baltimore, MD. MIM Number: 609248. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Rodriguez, C.I., et al. 2007. Disruption of the ubiquitin ligase Herc4 causes defects in spermatozoan maturation and impaired fertility. *Dev. Biol.* 312: 501-508.
5. Mählknecht, U., et al. 2009. Chromosomal characterization and localization of the NAD⁺-dependent histone deacetylase gene sirtuin 1 in the mouse. *Int. J. Mol. Med.* 23: 245-252.
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CHROMOSOMAL LOCATION

Genetic locus: HERC4 (human) mapping to 10q21.3.

PRODUCT

Herc4 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 Herc4 shRNA Plasmid (h): sc-90436-SH and Herc4 shRNA (h) Lentiviral Particles: sc-90436-V as alternate gene silencing products.

For independent verification of Herc4 (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-90436A, sc-90436B and sc-90436C.

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

Herc4 siRNA (h) is recommended for the inhibition of Herc4 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Herc4 gene expression knockdown using RT-PCR Primer: Herc4 (h)-PR: sc-90436-PR (20 μ l). 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.