

nardilysin siRNA (h): sc-41550

BACKGROUND

Aspartyl, serine, thiol and metalloenzyme proteases can be endoproteases, which activate protein precursors by cleavage at basic residues. Human nardilysin, also designated N-arginine dibasic convertase, NRD1 or NRD convertase, is a 1,147 amino acid metalloendopeptidase that cleaves propeptide and proprotein substrates at the amino-terminus of arginine residues in dibasic moieties. The nardilysin gene maps to chromosome 1p32.3 and is expressed as a 3.6-kb transcript primarily in adult heart, skeletal muscle and testis. In the testis, nardilysin appears to be restricted to germ cells. As a member of the insulinase family, nardilysin is a specific receptor for heparin-binding epidermal growth factor-like growth factor (HB-EGF) that modulates HB-EGF-induced cell migration via ErbB1. Nardilysin exhibits a significant degree of similarity to insulinase and to two yeast processing enzymes, Axl1 and Ste2. Defects in the gene encoding nardilysin are linked to inherited neuromuscular disorders.

REFERENCES

1. Pierotti, A.R., et al. 1994. N-arginine dibasic convertase, a metalloendopeptidase as a prototype of a class of processing enzymes. *Proc. Natl. Acad. Sci. USA* 91: 6078-6082.
2. Chesneau, V., et al. 1996. NRD convertase: a putative processing endoprotease associated with the axoneme and the manchette in late spermatids. *J. Cell Sci.* 109: 2737-2745.
3. Hospital, V., et al. 1997. Human and rat testis express two mRNA species encoding variants of NRD convertase, a metalloendopeptidase of the insulinase family. *Biochem. J.* 327: 773-779.
4. Fumagalli, P., et al. 1998. Human NRD convertase: a highly conserved metalloendopeptidase expressed at specific sites during development and in adult tissues. *Genomics* 47: 238-245.
5. Online Mendelian Inheritance in Man, OMIM™. 1998. Johns Hopkins University, Baltimore, MD. MIM Number: 602651. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim>
6. Nishi, E., et al. 2001. N-arginine dibasic convertase is a specific receptor for heparin-binding EGF-like growth factor that mediates cell migration. *EMBO J.* 20: 3342-3350.

CHROMOSOMAL LOCATION

Genetic locus: NRD1 (human) mapping to 1p32.3.

PRODUCT

nardilysin 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 nardilysin shRNA Plasmid (h): sc-41550-SH and nardilysin shRNA (h) Lentiviral Particles: sc-41550-V as alternate gene silencing products.

For independent verification of nardilysin (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-41550A, sc-41550B and sc-41550C.

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

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

nardilysin (A-6): sc-137199 is recommended as a control antibody for monitoring of nardilysin 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 nardilysin gene expression knockdown using RT-PCR Primer: nardilysin (h)-PR: sc-41550-PR (20 μ l, 410 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.

PROTOCOLS

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